



# ***JPRS Report***

## **Science & Technology**

***USSR: Life Sciences***

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# Science & Technology

## USSR: Life Sciences

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**Enhanced Efficiency of Remote Grain Hybridization by Embryo Culture**

917C0246C Moscow TSITOLOGIYA I GENETIKA  
in Russian Vol 24 No 5, Sep-Oct 90 (manuscript  
received 13 Mar 89) pp 25-28

[Article by V. P. Bannikova, P. D. Maystrov, Ye. A. Barabanova, Ye. A. Kravets, and G. S. Kolyuchaya, Institute of Botany, Ukrainian SSR Academy of Sciences, Kiev; Mironovka Scientific Research Institute of Wheat Selection and Breeding]

UDC 581.143:582.542.1

[Abstract] Trials were conducted on regeneration of wheat-rye hybrid plants using embryo rescue and callus induction from the hybrid embryo. Specifically, the study involved the cross combinations of Mironovskaya 808 wheat with the winter rye varieties Malysh 72 and Chulpan. The callus induction technique yielded 2.24 plants per embryo, demonstrating its superiority over embryo rescue which provided 0.72 hybrid plants per embryo. The high yield with the former methodology was sufficient to overcome the disadvantage of 19 percent incidence of amphidiploid and aneuploid plants. Figures 1; tables 2; references 9: 5 Russian, 4 Western.

**Selection of Yellow-Seed Varieties of Surepitza Grass and "Type 000" Rape**

917C0369 Moscow DOKLADY VSESOYUZNOY  
ORDENA LENINA I ORDENA TRUDOVOGO  
KRASNOGO ZNAMENI AKADEMII  
SELSKOKHOZYAYSTVENNYKH NAUK IMENI V. I.  
LENINA in Russian No 10, Oct 90 pp 25-28

[Article by V. I. Shpota and Ye. B. Bochkareva; All-Union Scientific Research Institute of Oleaginous Crops imeni V. S. Pystovoyt]

UDC 633.853.492+633.853.494:631.52

[Abstract] Development of domestic varieties of surepitza grass and "type 000" rape, performed at the Central Experimental Base of the All-Union Scientific Research Institute of Oleaginous Crops in 1978-1989, employed individual seed selection, inbreeding and

induced mutagenesis. Collection samples of the All-Union Scientific Research Institute of Plant Growing provided starting material: I-427773 for surepitza grass consisting of glaucous (60 percent) seeds and yellow (40 percent) seeds and k-4984 for rape. The procedures produced promising yellow-seed, non-erucic seeds with glucosinolates level in the seeds 2 to 3 times lower than that in the standard variety. Oil content of the seeds exceeded that of the standard variety by 2.2-4.0 percent and the yield was 13-23 percent higher than that of the mean value of the experiment. They differed from the standard glaucous seed variety by the low amount of husk of the seeds and the resistance to lodging. The rape and surepitza grass varieties can provide the fat and oil industry lightly-pigmented food vegetable oil and the combination fodder industry a source of improved fodder concentrate. References 8: 4 Russian, 4 Western.

**Secondary Metabolites in *Pyricularia Oryzae***

917C0409B Tashkent KHIMIYA PRIRODNYKH  
SOYEDINENIY in Russian No 6, Nov-Dec 90  
(manuscript received 28 Apr 89, after revision  
24 May 90) pp 811-818

[Article by S. I. Sviridov and B. S. Yermolinskiy; All-Union Scientific Research Institute of Phytopathology, Moscow Oblast]

UDC 547.564.3:581.192:632.488

[Abstract] The following *o*-nitrophenol derivatives were isolated from cultured fluid from the *Pyricularia oryzae* Cav. VG-345 deuteromycete: 3-nitro-4-hydroxy phenyl acetic acid (I), 3-nitro-4-hydroxy benzyl alcohol, I and 2-(3-nitro-4-hydroxy phenyl)-ethanol (III), N-(3-nitro-4-hydroxy-phenyl-ethyl)-acetamide, and pyricularamide (a new compound discovered and named by the authors). The authors proposed that pyricularamide has a peptide nature and assigned diketo piperazine, nitrotyrosine, and proline structures to it. All the compounds were isolated from natural sources for the first time. Compounds I and III exhibited moderate growth inhibiting activity (70 + 7 and 45 + 7 percent, respectively) on rice seedlings at concentrations of 60 µg/ml. Compound III caused root deformation. Figures 1; references 17: 3 Russian, 14 Western.

**Control of Mitochondrial Energy Metabolism by Biogenic Amines**

917C0226A Moscow VOPROSY MEDITSINSKOY  
KHIMII in Russian Vol 36 No 5, Sep-Oct 90  
(manuscript received 16 Jan 89) pp 18-21

[Article by A. Ye. Medvedev, Institute of Biological and Medical Chemistry, USSR Academy of Medical Sciences, Moscow]

UDC 612.014.2:576.347].014.46:[615.357:577.175.823

[Abstract] Studies were conducted on rat hepatic mitochondria to determine the effects of biogenic amines on mitochondrial (MT) energy metabolism in the face of monoamine oxidase (MAO) inhibition. Incubation of the MT membranes with the amines in the presence of MAO inhibitors (1  $\mu$ M chlorgilin + 1  $\mu$ M deprenyl) led to inhibition of rotenone-insensitive NAD-H cytochrome C reductase, succinate cytochrome C reductase, and succinate dehydrogenase. The inhibitory efficacy of the amines ranked as follows: 3 mM serotonin > 2.4 mM phenylethylamine > 30  $\mu$ M spermine > 3 mM tryptamine > 3 mM tyramine > 3 mM dopamine. These observations demonstrate that the amines inhibited enzymes of the external and internal membranes through changes in the surface charge of the membranes. In view of the extremely low *in vivo* concentrations of the biogenic amines such mechanisms could only prevail in pathologic conditions or under the influence of drugs acting as MAO inhibitors when the concentration of the amines may be elevated 2-fold or so. In such cases the effects of biogenic amines on mitochondrial metabolism could clearly have pathogenetic significance. Figures 2; tables 1; references 24: 10 Russian, 14 Western.

**Effects of Trichothecene Mycotoxin Deoxynivalenol (Vomitoxin) on Calcium Homeostasis and Vitamin D Metabolism and Receptors in Rats**

917C0226B Moscow VOPROSY MEDITSINSKOY  
KHIMII in Russian Vol 36 No 5, Sep-Oct 90  
(manuscript received 28 Feb 89) pp 26-29

[Article by I. N. Sergeyev, L. V. Kravchenko, N. M. Piliya, A. B. Batukhanov, V. S. Sobolev, Ye. E. Kuzmina, L. M. Yakushina, V. B. Spirichev, and V. A. Tutelyan, Institute of Nutrition, USSR Academy of Medical Sciences, Moscow]

UDC 616-099-02:[612.263:633.11:632.4]-  
07:616-008.924.1]-092.9

[Abstract] Toxicity testing was conducted with vomitoxin in terms of calcium and vitamin D metabolism. Studies on 50 g male Wistar rats treated with 10 mg/kg/day of vomitoxin per os for seven days demonstrated that such animals developed moderate hypocalcemia as a result of diminished intestinal absorption of calcium, although bone density was not affected. Concomitantly,

alkaline phosphatase activity of the blood and intestinal mucosa decreased. Other pertinent changes included a 40 percent reduction in blood levels of 25-OHD, a vitamin D metabolite, and a 30 percent reduction in the activity of hepatic 1- and 24-hydroxylases of 25-OHD<sub>3</sub>. However, renal receptors for 1,25(OH)<sub>2</sub>D<sub>3</sub> and 24,25(OH)<sub>2</sub>D<sub>3</sub> remained unaffected. Such changes did not occur in animals maintained on a diet incorporating 0.25 mg/kg of vitamin D. Accordingly, these observations lead to the conclusion that vitamin D deficiency is a significant factor in vomitoxin toxicity. Tables 2; references 13: 7 Russian, 6 Western.

**Glycyrrhizic Acid Salts: Antioxidant Properties and Action on Hepatic Microsomal Monooxygenase System**

917C0226C Moscow VOPROSY MEDITSINSKOY  
KHIMII in Russian Vol 36 No 5, Sep-Oct 90  
(manuscript received 1 Mar 89) pp 29-31

[Article by M. A. Abdugafurova, V. S. Li, M. P. Sherstnev, T. B. Atanayev, A. Sh. Isamukhamedov, and G. I. Bachmanova, Scientific Research Institute of Physicochemical Medicine, RSFSR Ministry of Health; Second Moscow Medical Institute imeni N. I. Pirogov, Moscow]

UDC  
615.322:547.918].015.4:[612.351.11:577.152.199.2

[Abstract] Chemiluminescence quenching studies on microsomes derived from outbred 100-120 g male rats led to the demonstration that sodium and potassium salts of glycyrrhizic acid (NaG, Na<sub>3</sub>G, K<sub>3</sub>G) were capable of forming type I complexes with cytochrome P-450. The net effect consisted of stabilization of the reduced hemoprotein. Additionally, *in vivo* studies on the intraperitoneal administration of Na<sub>3</sub>G (2 mg/100 g/day for three days) showed that it led to the induction of cytochrome P-450. These findings suggest that such salts may have therapeutic applications in reversing hepatic toxicity. Figures 3; tables 2; references 17: 4 Russian, 13 Western.

**Multiple Forms of Cerebral Monoamine Oxidase and Experimental Catatonia in Rats**

917C0226D Moscow VOPROSY MEDITSINSKOY  
KHIMII in Russian Vol 36 No 5, Sep-Oct 90  
(manuscript received 17 Mar 89) pp 32-34

[Article by N. S. Kamyshanskaya, V. Z. Gorkin, and N. N. Voytenko, Institute of Biological and Medical Chemistry, USSR Academy of Medical Sciences, Moscow; Institute of Cytology and Genetics, Siberian Department, USSR Academy of Sciences, Novosibirsk]

UDC 616.89-008.431-092.9-07:616.831-  
008.931:577.152.143.042.2

[Abstract] An analysis was conducted on the brain stem activities of monoamine oxidase isozymes in male Wistar rats predisposed to catatonia. The isozymes were

separated by affinity chromatography on AN-Sepharose columns into four fractions (I, II $\alpha$ , II $\beta$ , III) and analyzed for deamination of serotonin, tyramine and 2-phenylethylamine. Comparison of isozyme levels in control and catatonia-prone rats showed an absence of significant differences, although fraction II $\alpha$  tended to be higher in the latter animals and fraction III in the former. However, the rates of tyramine and 2-phenylethylamine deamination by isozymes II $\beta$  and III were much greater ( $P < 0.05$ ) in the catatonia-prone rats than in the control animals. Conversely, the rate of serotonin deamination by isozyme III in the catatonia-prone animals was significantly below that seen in the control rats. These observations may indicate pathogenetically important differences, which are not evident if such studies are performed with unfractionated monoamine oxidase. Figures 2; tables 2; references 15: 7 Russian, 8 Western.

#### Prevention of Acute T-2 Mycotoxicosis by Dietary Selenium

917C0226E Moscow VOPROSY MEDITSINSKOY  
KHIMII in Russian Vol 36 No 5, Sep-Oct 90  
(manuscript received 19 Apr 90) pp 36-38

[Article by L. V. Kravchenko, Ye. E. Kuzmina, L. I. Avrenyeva, and V. A. Tutelyan, Institute of Nutrition, USSR Academy of Medical Sciences, Moscow]

UDC 546.23:615.9:577.15.04

[Abstract] Male Wistar rats with an initial weight of 50 g were used in an assessment of the therapeutic potential of selenium in T-2 mycotoxicosis. Experimental animals were maintained for six weeks on a selenium-enriched diet (0.5-2.5 mg/kg of feed), following which control and experimental rats were treated intragastrically with 3.8 mg/kg T-2 toxin in 1 percent ethanol. Biochemical and clinical monitoring of T-2 intoxication showed a reduction in mortality from 47 percent in unprotected rats to 20 percent in those on the 2.5 mg/kg selenium diet. The reduction in mortality was accompanied by a milder course of mycotoxicosis and less pronounced biochemical perturbations. Specifically, in the experimental group hydroxylation of T-2 to 3'-hydroxy-T-2 was attenuated and elevated UDP-glucuronosyl transferase activity indicated more efficient metabolic conjugation of T-2 and its metabolites with glucuronic acid. On balance, the biochemical studies showed that dietary selenium favored a metabolic shift in the biotransformation of T-2 toward more efficient conjugation and elimination from the body. Absence of changes in hepatic glutathione peroxidase activity indicates that mitigation of T-2 toxicity was unrelated to antioxidant mechanisms. Figures 1; tables 2; references 15: 2 Russian, 13 Western.

#### Calcium Metabolism Disorders in T-2 Toxicosis: Endocrine and Vitamin D Factors

917C0226F Moscow VOPROSY MEDITSINSKOY  
KHIMII in Russian Vol 36 No 5, Sep-Oct 90  
(manuscript received 31 Oct 89) pp 45-48

[Article by I. N. Sergeyev, N. M. Piliya, V. A. Tutelyan, and V. B. Spiridov, Institute of Nutrition, USSR Academy of Medical Sciences, Moscow]

UDC 616.152.41-008.64-092.9-02:615.918:582.282]-  
06:616.154:577.161.2

[Abstract] An analysis was conducted on the effects of T-2 mycotoxin on the metabolism and receptor binding of cholecalciferol (vitamin D). The experimental model system consisted of 50 g male Wistar rats kept on a cholecalciferol-deficient diet for four weeks; one to five days before sacrifice the animals were treated with 40 IU/day of cholecalciferol and/or with 0.54 mg/kg/day (1/7 LD<sub>50</sub>) of T-2 toxin. The metabolic studies showed that T-2 treatment markedly attenuated hepatic transformation of cholecalciferol to 25-hydroxycholecalciferol. In addition, target tissues became far less susceptible to the vitamin as is evident in sharply reduced binding of 1,25-dihydroxycalciferol to renal receptors. Concomitantly, concentrations of renal cAMP and serum parathyroid hormone rose markedly. These findings provide further support that T-2 toxin effects on calcium metabolism are based on interference with cholecalciferol metabolism. In view of the significance of 1,25-dihydroxycalciferol in cell differentiation and proliferation, immune response and hormone production and secretion, it appears that the full range of T-2 pathogenetic mechanisms still remains to be elucidated. Figures 1; tables 3; references 8: Russian.

#### Effects of Enteric Adsorbents on Lipid Peroxidation and Antioxidant Properties of Hepatic and Blood Lipids in Combined Heat Burns

917C0226G Moscow VOPROSY MEDITSINSKOY  
KHIMII in Russian Vol 36 No 5, Sep-Oct 90  
(manuscript received 20 Nov 89) pp 61-63

[Article by A. S. Seylanov and B. M. Nurkhanov, Scientific Research Institute of Medical Radiology, USSR Academy of Medical Sciences, Obninsk]

UDC 616-001.17-085.246.2-032:611.33/  
.34]-036.8-07:616.36-008.939.15

[Abstract] Experimental therapeutic trials were conducted with several gastrointestinal adsorbents on rats subjected to heat burns. Male Wistar rats, 200-240 g in weight, were treated intragastrically with fibrous carbon adsorbent (0.3 g/kg), polymethylsiloxane (4 g/kg), or piperidol homo- or copolymer (0.5 g/kg) 1 and 24 h after a heat burn over 15 percent of the body surface from a

quartz-halogen lamp flash. Biochemical studies on the liver showed that each preparation attenuated hepatic lipid peroxidation and promoted recovery of endogenous GSH. The hepatoprotective effects were attributed to adsorption and accelerated elimination of endogenous toxins arising from tissue damage. The more moderate rise in plasma levels of lipid peroxidation products varied with the type of adsorbent, and also was attributed to enhanced antioxidant properties of the plasma

lipids. Consequently, these observations also indicated that the adsorbents differed somewhat in their mechanisms of action. In the final analysis, in addition to demonstrating the efficacy of gastrointestinal adsorbents in limiting heat burn-induced pathology, the data also indicated that hepatic levels of malonic dialdehyde, a product of lipid peroxidation, can be used as a marker of therapeutic success. Tables 2; references 13: 11 Russian, 2 Western.

**Effect of  $\alpha$ -Latrotoxin on Glutamate Accumulation and Release in Rat Brain Synaptosomes**

917C0357A Moscow BIOLOGICHESKIYE  
MEMBRANY in Russian Vol 7 No 10, Oct 90  
pp 1013-1021

[Article by S. I. Zharikov, A. D. Zharikova, V. I. Popov, V. N. Pashkov, and Ye. V. Grishin, Institute of Biological Physics, USSR Academy of Sciences, Pushchino, Moscow Oblast; Pushchino Branch, Institute of Bioorganic Chemistry imeni M. M. Shemyakin, USSR Academy of Sciences, Pushchino, Moscow Oblast; Institute of Bioorganic Chemistry imeni M. M. Shemyakin, Moscow]

UDC 577.175.82:591.145.2

[Abstract] LT or  $\alpha$ -Latrotoxin—a high-molecular protein from the venom of the *Latrodectus mactans tredecimguttatus* spider—is a neurotoxin that produces a pre-synaptic effect and stimulates massive release of a number of neuromediators in the synapses of the peripheral and central nervous systems. Available data suggest that the effect produced by LT is nonselective in terms of neuromediators and consists in amplification of the exocytosis of such compounds from nerve endings. The mechanism of the accelerated exocytosis has yet to be studied, and the same is true of the effect of LT on the systems for the release and  $\text{Na}^+$ -dependent uptake of glutamate. In light of that, as well as the fact that glutamate is widely used by excitatory neurons of the central nervous system, the researchers here studied the effect of LT on glutamate accumulation and release in rat brain synaptosomes. They found that, in the context of binding kinetics, specific binding sites on the synaptosomes for LT were completely filled within two minutes. LT diminished the amount of glutamate accumulating in the synaptosomes over a five minute period. Within the first two minutes of incubation with glutamate, however, LT had no effect on  $\text{Na}^+$ -dependent accumulation of glutamate. As for effect on release, LT accelerated the release of the accumulated glutamate within two minutes of its addition to the incubation medium. Electron microscopy of the synaptosomes indicated that the synaptosomes constituted the greater portion of the preparations under study. Synaptic vesicles within the synaptosomes varied in size, shape, and distribution. Many of the synaptosome profiles contained mitochondria, with 30 percent of the profiles having so-called active zones, i.e., bound with postsynaptic elements. After exposure to LT, the synaptosomes exhibited morphological changes, chief among them the

shift of the synaptic vesicles to the active zones. Figures 8, references 22: 4 Russian, 18 Western.

**Protonation-Deprotonation Reaction of Nigericin on the Surface of a Bilayer Lipid Membrane is a Limiting Stage of Ion Transport**

917C0357B Moscow BIOLOGICHESKIYE  
MEMBRANY in Russian Vol 7 No 10, Oct 90  
pp 1037-1044

[Article by Yu. N. Antonenko, O. N. Kovbasnyuk, and L. S. Yaguzhinskiy, Interdepartmental Problem Scientific-Research Laboratory of Molecular Biology and Bioorganic Chemistry imeni A. N. Belozerskiy, Moscow State University imeni M. B. Lomonosov]

UDC 577.352.465

[Abstract] The reaction rate of enzyme deprotonation on a membrane surface has been shown to increase with elevation of buffer concentration during the operation of the proton pump of bacteriorhodopsin from *Halobacterium halobium*. Increasing the buffer concentration also boosts the rate of transport of hydrogen ions between reagents in aqueous phase and reagents on a membrane surface. Other research has shown that when cations are transported through a gramicidin channel, the stage of cation diffusion in the area of contact between the mouth of the channel and the aqueous solution is a limiting stage. That and other findings suggest that reactions involving addition/detachment of protons on a membrane surface can take place at a lower rate than that associated with the transmembrane transport of those ions. In the work reported here, the researchers conducted a two-part study of reactions involving addition/detachment of hydrogen ions on the surface of a bilayer lipid membrane (BLM) when ion transport was induced. They examined the effect of the buffer volume of solutions on nigericin-induced cation transport through BLMs. In the process, they developed a modification of a technique for recording nonelectrogenic fluxes of hydrogen ions. The technique makes it possible to perform measurements in the absence of a local pH gradient in the premembrane layers. Figures 5; references 23: 3 Russian, 20 Western.

**Cationic-Polypeptide-Induced Formation of Nonbilayer Structures in Phospholipid Membrane**

917C0357C Moscow BIOLOGICHESKIYE  
MEMBRANY in Russian Vol 7 No 10, Oct 90  
pp 1045-1055

[Article by S. E. Gasanov, B. A. Salakhutdinov, and T. F. Aripov, Institute of Bioorganic Chemistry imeni A. S. Sadykov, UzSSR Academy of Sciences, Tashkent]

UDC 577.353.335

[Abstract] Many cell-membrane and extracellular proteins bind reversibly with biological membranes, and the binding plays a prominent role in the functioning of water-soluble proteins in living organisms. Membrane binding is apparently a common stage in the transmembrane transport of proteins. Certain water-soluble proteins bind with membranes via hydrophobic "anchor" groups that are capable of interacting with the hydrophobic part of a lipid bilayer. A necessary condition for the translocation of the protein through the membrane is the coupling of that process with protein-synthesis processes. Techniques involving magnetic resonance and triplet probes were used here to study the interaction between, on the one hand, two homologous cytotoxins,  $V_{c1}$  and  $V_{c5}$ , extracted from the venom of the Central Asian cobra and, on the other, phospholipid membranes of varying composition. Those cytotoxins were chosen because they exhibit membrane-active properties in aqueous solutions and because lipid-bilayer localization has been determined for them, as have amino acid sequence and molecule size and shape. The researchers found that the two cytotoxins modify a phosphatidyl membrane that contains negatively charged phospholipids. Two types of nonbilayer structures are formed: (1) structures with a fast isotropic phospholipid motion, and (2) structures with a wide angular distribution of phospholipid molecular axes, whose mobility is restricted. The formation of nonbilayer structures is largely determined by the molecular distribution of cytotoxins in the lipid and aqueous phases of the liposomes. Figures 6; references 51: 24 Russian, 27 Western.

#### Selective Hydrophobization of IgG Fab' Fragments and Formation of Langmuir Films From Them

917C0357D Moscow *BIOLOGICHESKIYE MEMBRANY* in Russian Vol 7 No 10, Oct 90  
pp 1081-1085

[Article by I. V. Turko, I. A. Piluleva, and V. V. Yerokhin, Institute of Bioorganic Chemistry, BSSR Academy of Sciences, Minsk; Institute of Crystallography imeni A. V. Shubnikov, USSR Academy of Sciences, Moscow]

UDC 577.112.389.4

[Abstract] The creation of complex protein structures ordered at the molecular level is the focus of much attention because they are expected to be used in the future to construct new types of biosensors and bioelectronic devices. The technology for producing Langmuir films, which consists in the sequential transport of single layers of organic molecules from a water surface to a solid substrate, makes it possible to create three-dimensional single-layer and multilayer structures that have given optical and electrical properties. Immunoglobulin G is a water-soluble protein, and its use in the

production of Langmuir films without preliminary modification has drawbacks such as the difficulty in predicting the orientation of the molecules in the film. In light of that, the researchers here synthesized IgG Fab' fragments selectively hydrophobized, produced Langmuir films from them, and found the films to have good antigen-binding properties. Figures 4; references 7: 1 Russian, 6 Western.

#### Comparative Study of Single Layers of Proteins of Photosynthetic Reaction Centers From Green Filament and Purple Bacteria

917C0357E Moscow *BIOLOGICHESKIYE MEMBRANY* in Russian Vol 7 No 10, Oct 90  
pp 1086-1092

[Article by N. A. Kalabina, S. Yu. Zaytsev, M. A. Kutuzov, A. S. Zolotarev, Ye. P. Lukashev, A. A. Kononenko, N. G. Abdulayev, and V. P. Zubov, Institute of Bioorganic Chemistry imeni M. M. Shemyakin, USSR Academy of Sciences, Moscow; Moscow State University imeni M. V. Lomonosov]

UDC 577.352.2+577.122

[Abstract] A comparative study was performed of the structure and properties of reaction centers of *Chloroflexus aurantiacus* and *Rhodobacter sphaeroides* in a single layer on the surface of a water/air interface, and optimum conditions for the formation of single layers and for the transport to a solid substrate were determined. The researchers found that both reaction centers formed stable single layers at a transition pressure of 31-31 mN/m, which is close to the surface tension in bilayer phospholipid membranes. The areas of the protein molecules of the reaction centers in the single layers were similar. But area in *Rh. sphaeroides* was heavily dependent on subphase ionic strength variation, which had little effect on area in *C. aurantiacus*. Optimum conditions for the formation of the single layers were provided by a 10 mM phosphate buffer, pH 7.0-8.0, at 20°C. Figures 8; references 13: 3 Russian, 10 Western.

#### Synthesis and Surface-Active Properties of 1-, 3-, and 1,3-Substituted Pyrroles

917C0357F Moscow *BIOLOGICHESKIYE MEMBRANY* in Russian Vol 7 No 10, Oct 90  
pp 1093-1096

[Article by S. L. Vorobyeva, N. V. Bezobrazova, and P. S. Sotnikov, Scientific Research Institute of Physical Problems imeni F. V. Lukin, Moscow]

UDC 547.741.057+539.216.2

[Abstract] Pyrrole-based polymers are known to be highly conductive, with conductivity rising as high as 600 ohm<sup>-1</sup>/cm<sup>-1</sup> when the polymers are doped with iodine. Electrochemical and chemical techniques of polymerization are of great interest to many researchers

in the creation of morphologically and structurally advanced thin layers on dielectric substrates, and one promising technique is the production of Langmuir-Blodgett films based on pyrrole derivatives. The simultaneous insertion of substituents into positions 1 and 3 exerts a substantial effect on monomer reactivity in polymerization and on molecule orientation in single layers. That prompted the researchers to synthesize surface-active monopyrroles and dipyrroles with substituents at positions 1, 3, and 1.3 and to study the properties of the compounds they produced. They demonstrated that stable single layers and Langmuir-Blodgett films can be produced on the basis of 3-octadecanoylpyrrole, 1-methyl-3-octadecanoylpyrrole, 1-acetyl-3-octadecanoylpyrrole, 1,5-dioxo-1,5-bis(3-octadecanoylpyrrole-1)-yl-pentane, and 1,6-(3-octadecanoylpyrrole-1)-2,4-diheptene. Figures 5; references 3: Western.

**Study of the Kinetics of Photocoloration-Decoloration of Amphiphilic Indolinonaphthooxazine Derivatives in Multilayer Langmuir-Blodgett Films**

917C0357G Moscow BIOLOGICHESKIYE  
MEMBRANY in Russian Vol 7 No 10, Oct 90  
pp 1097-1104

[Article by A. M. Vinogradov, Ye. I. Kuznetsov, I. R. Mardaleyshvili, V. M. Anisimov, A. V. Lyubimov, M. V. Alfimov, V. S. Marevtsev, M. I. Cherkashin, and V. I. Bolshakov, Institute of Chemical Physics, USSR Academy of Sciences, Moscow]

UDC 577.352.2

[Abstract] The fact that the photochromic properties of spiroindolinonaphthooxazines in single layers on the surface of a water/air interface and in multilayer Langmuir-Blodgett films are little studied prompted the researchers here to synthesize those compounds with a structure such that  $R = C_{18}H_{37}$  (I) and  $R = CH_3$  (II) in a study of the conditions attending the production of photochromic Langmuir-Blodgett based on amphiphilic derivatives. Absorption spectra of dyed merocyanine forms in the Langmuir-Blodgett films were found to shift to the short-wave region and broaden. Decoloration kinetics for those forms obeyed the first-order law in alcohol and in water. In studying  $\pi$ -A-isotherms for mixtures of spironaphthooxazines and stearic acid in various molar ratios, the researchers found the area per molecule of spironaphthooxazine to be 68 angstroms. Decoloration kinetics for the merocyanine forms in the Y Langmuir-Blodgett films based on a 1:6 mixture of spironaphthooxazine and stearic acid are described in the context of distributed reactivity. Figures 5; references 14: 2 Russian, 12 Western.

**Conducting Langmuir-Blodgett Films of Complexes With Charge Transfer of Surface-Active Derivatives of DMTTF and TCNQ: Electrical Properties and Structure as a Function of Composition**

917C0357H Moscow BIOLOGICHESKIYE  
MEMBRANY in Russian Vol 7 No 10, Oct 90  
pp 1105-1110

[Article by L. A. Galchenkov, S. N. Ivanov, F. Ya. Nad, V. P. Chernov, T. S. Berzina, and V. I. Troitskiy, Institute of Radioengineering and Electronics, USSR Academy of Sciences, Moscow; USSR Ministry of Electronics Industry, Moscow]

UDC 538.975

[Abstract] Conducting Langmuir-Blodgett films are coming under intense scrutiny today because of their possible use in molecular electronics. The work reported here focuses on a study of conducting films of complexes with charge transfer. The films are prepared from a mixture of  $C_{16}H_{33}$ -TCNQ (I) and  $C_{17}H_{35}$ -DMTTF (II). The researchers studied  $(I)_{1-x}(II)_x$  films in which  $x$  ranged from 0.5 to 0.71. Temperature dependences of the conductivity were measured in the range 140-320 K; the Hall effect was also measured, at room temperature. The temperature dependences exhibited a maximum at  $T_{max}$  of approximately 300 K, with  $\sigma$  approximately  $0.1-1.0 \text{ ohm}^{-1}/\text{cm}^{-1}$ . The activation nature of the conductivity was observed at  $T < T_{max}$ , with energy of activation 0.12-0.18 eV for various mixtures. At  $T > T_{max}$ , conductivity dropped sharply. Figures 4; references 6: Western.

**Intermembrane Exchange by Lipids, Induced by Cobra Venom Cytotoxins**

917C0393A Moscow BIOFIZIKA in Russian Vol 35  
No 6, Nov-Dec 90 (manuscript received 11 Jan 88)  
pp 958-962

[Article by S. E. Gasanov, T. F. Aripov and B. A. Salakhutdinov; Institute of Bioorganic Chemistry; UzSSR Academy of Sciences; Tashkent]

[Abstract] Studies of the interaction of cobra venom cytotoxin  $V_{c5}$  with model membranes from phosphatidylcholine and phosphatidic acid and intermembrane mixing of lipids employed  $^1\text{H}$ -NMR methods, ESR spin probes and microcalorimetry in a system of oriented multibilayers. Intermembrane mixing of lipids, induced by the cytotoxin was studied by use of differential scanning microcalorimetry. Interaction of the cytotoxin with the membrane modified the bilayer with formation of non-bilayer structures with wide angle distribution of orientations of molecular axes of the lipids. This disturbed the integrity of the membranes, increased the membrane permeability and stimulated intermembrane exchange by the lipids. Figures 4; references 19: 9 Russian, 10 Western.

**Role of Melanin in Regulating Free-Radical Processes in Pigmental Epithelium of Eye During Acute Hypoxia**

917C0393B Moscow *BIOFIZIKA in Russian* Vol 35  
No 6, Nov-Dec 90 (manuscript received 30 Nov 89)  
pp 977-980

[Article by N. M. Magomedov, A. I. Dzhaferov and E. Yu. Yusifov; Institute of Physiology imeni A. I. Kareyev and Scientific-Industrial Association "Biotekh"; Academy of Sciences; AzSSR; Baku]

UDC 577.3

[Abstract] Experiments performed on the eyes of pigmented and albino guinea pigs involved development of acute hypoxia in an airtight 0.12 m<sup>3</sup> chamber for 1.5 hours by constantly pumping through a mixture of gases (5 percent O<sub>2</sub> and 95 percent N<sub>2</sub>) through the chamber produced reoxygenation. Decapitation of the animals immediately after hypoxia and hypoxia with reoxygenation, removal of the retina and pigmental epithelium and the accompanying vascular layer from the optic cup and all further operations were performed in daylight. Reaction of the retina to acute hypoxia and reoxygenation did not depend upon the presence of melanin in the pigmental epithelium of the eye. Hypoxia increased the level of hydroperoxides and malonic dialdehyde in the retina of both pigmented and albino animals. Reoxygenation produced an even greater accumulation of products of peroxide oxygenation of lipids in retinas of both pigmented and albino animals. Reoxygenation decreased below the norm the quantity of malonic aldehyde in the pigmented epithelium of melanin-containing animals, but not in albinos. Acute hypoxia increased the electron spin resonance-absorption of the pigmented epithelium. Reoxygenation greatly decreased the electron spin resonance signal intensity. Preliminary introduction of sodium selenite against this background increased the ERS-absorption intensity of the pigmented

epithelium 2.5 times with return to the control level after 20 minutes of reoxygenation. It was assumed that, during hypoxia with subsequent reoxygenation, the melanin-protein granules promote regulation of peroxide oxidation of the lipids only in the pigmental epithelium and not in the other layers of the retina. This differs greatly from cases connected with the effect of strong illumination, described in the literature. Figures 2; references 15: 9 Russian, 6 Western.

**Behavior of Magnetic Particles of Metallic Iron in Animal Organism**

917C0393C Moscow *BIOFIZIKA in Russian* Vol 35  
No 6, Nov-Dec 90 (manuscript received 12 Nov 89)  
pp 985-988

[Article by M. M. Shabarchina, A. I. Tsapin, A. G. Malenkov and A. F. Vanin; Institute of Chemical Physics; Academy of Sciences USSR; Moscow]

[Abstract] A comparison of the behavior of large and small particles (5-8  $\mu$ m and less than 1  $\mu$ m) of metallic iron in the animal organism involved experiments on 100 white mongrel 18-20 g mice and 10 90-100 g rats. Electron spin resonance studies provided a good assessment of the phase state of the iron particles in the organism (ferromagnetic, antiferromagnetic and ionic). Intraperitoneal administration of ferromagnetic 5-8  $\mu$ m particles to the mice produced capsulation and preservation in the organism for at least one month. Activation of peroxide oxidation of lipids occurred on the third or fourth day after entry of these particles with a 2-fold increase of free iron level in the animals' liver. Intraperitoneal administration to mice of particles less than 1  $\mu$ m caused rapid death. Intravenous injection of ferromagnetic 5-8  $\mu$ m particles to rats caused their accumulation in the liver, lungs and spleen due to trapping of these particles by macrophages. The particles were transformed within two weeks and disappeared from these tissues. Figures 2; references 8: 5 Russian, 3 Western.

**Incidence of Chromosomal Mutations in Bone Marrow Cells From Wild Mice Captured in Areas With High Pesticide Use**

917C0246A Moscow TSITOLOGIYA I GENETIKA  
in Russian Vol 24 No 5, Sep-Oct 90 (manuscript  
received 27 Mar 89) pp 10-13

[Article by P. Kh. Khalikov, Tashkent Medical Institute]

UDC 576.312.33.36:611-018.46:613.632

[Abstract] Cytogenetic studies on wild mice (21-24 g) captured in cotton fields heavily treated with pesticides in the Dzhizak Oblast showed that chromosomal rearrangements were present in 2.34 percent of the bone marrow cells, exceeding the incidence in laboratory-maintained mice almost 4-fold. The rearrangements were largely of the chromatid type, usually involving terminal deletions followed in frequency by intrachromosomal chromatic inversions. In addition, the percentage of polychromatophilic erythrocytes with micronuclei reached 10.22 percent in the experimental mice, almost 5-fold greater than the incidence in the bone marrow of control mice (1.97 percent). These findings demonstrated that ingested pesticides affect the genetic apparatus of mice. However, the frequency of both types of aberrations decreased to baseline after five months in a vivarium. Tables 1; references 8: 4 Russian, 4 Western.

**B Chromosome Polymorphism in *Rana Temporaria* L. Populations**

917C0246B Moscow TSITOLOGIYA I GENETIKA  
in Russian Vol 24 No 5, Sep-Oct 90 (manuscript  
received 10 Apr 89) pp 13-17

[Article by K. G. Yeliseyeva, M. V. Ploskaya, and A. M. Voytovich, Institute of Genetics and Cytology, Belorussian SSR Academy of Sciences, Minsk]

UDC 575.174.015.3+576.316.79

[Abstract] Studies in 1984-1986 on four populations of the frog *Rana temporaria* in two Minsk districts, a Minsk suburb, and at the Berezinskiy Biospheric Preserve revealed the effects of industrial pollution on the frequency of B chromosomes. The incidence of B chromosomes in frogs captured in a Minsk district with heavy industrial pollution was 6-fold higher than in frogs captured in the Berezinskiy Preserve, and 4- to 5-fold higher than in those captured in an unpolluted district and a Minsk suburb. In addition, some frogs captured in the district with high pollution had as many as seven B chromosomes, versus one to two in frogs from other locations. The data indicated that over 40 years in the district with high levels of pollution, the average number of B chromosomes per genome increased at the rate of 0.03 to 0.04 B chromosomes per generation. Tables 2; references 14: 7 Russian, 7 Western.

**Isolation and Description of Wheat Mutants Resistant to S-Aminoethylcysteine, Lysine, and Threonine**

917C0246E Moscow TSITOLOGIYA I GENETIKA  
in Russian Vol 24 No 5, Sep-Oct 90 (manuscript  
received 3 Apr 89) pp 37-41

[Article by V. A. Sidorov, V. V. Morgun, V. F. Logvinenko, and N. A. Matveyeva, Institutes of Botany and of Plant Physiology and Genetics, Ukrainian SSR Academy of Sciences, Kiev]

UDC 633.11:631.527

[Abstract] In order to improve wheat protein in terms of essential amino acid content, studies were undertaken on the induction and isolation of wheat mutants resistant to S-aminoethylcysteine, lysine, and threonine. Treatment of Kiyanka and Chayka seeds with chemical mutagens and subsequent cultivation of the embryos on selective media led to identification of S-aminoethylcysteine-resistant mutants with a frequency of  $4 \times 10^{-4}$  to  $1.2 \times 10^{-3}$ . Subsequent seed studies yielded resistant and susceptible segregants in the approximately anticipated ratios (5:3). Combined lysine + threonine resistance was obtained by callus induction from early embryos treated with a mutagen. The most promising wheat variety with this approach was Mironovskaya 808, and the least promising were the Chayka, Lutescens 7, and Novosibirskaya 67 varieties; Siete Cerros and Spring Mironovskaya varieties occupied intermediate positions. Figures 1; tables 3; references 11: Western.

**Air Pollution and Lung Cancer Morbidity of Urban Population**

917C0309A Moscow GIGIYENA I SANITARIYA  
in Russian No 12, Dec 90 (manuscript received  
5 Jun 89) pp 4-7

[Article by D. G. Zaridze and G. M. Zemlyanaya, Scientific Research Institute of Carcinogenesis, All-Union Scientific Oncology Center of the USSR Academy of Medical Sciences, Moscow]

UDC 616.24-006.6-02:614.715]-036.2-07

[Text] Lung cancer is now the most widespread form of cancer in the world, and it occupies first place in the structure of morbidity and mortality due to malignant tumors in all developed countries of the world, including the USSR. The high frequency of this disease and the significant difficulties in its early diagnosis and treatment predetermine the need for studying the etiology of lung cancer with the purpose of developing the methods of its prevention.

The role of environmental factors in the genesis of lung cancer is now universally recognized [3-5,8]. It has been shown that smoking, occupation and air pollution affect the risk of development of lung cancer. Smoking, with

which 70-95 percent of cases of this pathology are associated, has the most important significance in the etiology of this disease [8]. The proportion of lung cancer associated with occupational factors varies from 4 to 40 percent depending on the place and time of the research, and on the concentration of industrial enterprises in the research zone in a particular period of time [7].

Lung cancer is associated with pollution of the air by carcinogenic substances to a greater degree than other forms of malignant tumors.

The goal of this study was to determine the effect of two factors on lung cancer morbidity—air pollution and smoking—using 26 major cities of the Soviet Union as an example. The cities included in the study are located in dissimilar natural and climatic zones, and they possess different demographic characteristics, a different structure of industrial production, and a high amplitude of variations in lung cancer morbidity.

Updated data on lung cancer morbidity were collected for 1979-1983 on the basis of a review of reports (reporting form u-281) and verified data on reporting form No 61Zh. Age-sex and standardized indicators of lung cancer morbidity were calculated. The world standard was used as the standard [6]. Standardized lung cancer morbidity indicators for the cities in question, expressed per 100,000 population, exhibit significant differences: Male morbidity varies from 110.6 in Norilsk, 96.4 in Krivoy Rog and 95.5 in Magnitogorsk to 58.2 in Kursk, 54.1 in Voronezh and 51.8 in Yerevan; among women, the highest morbidity indicators are in Ust-Kamenogorsk—13.0, Magnitogorsk—12.7, Yerevan—12.4, and Rostov-on-Don—12.2, and the least are in Voronezh—7.9, in Vilnius—7.7, and in Ulyanovsk—6.2.

When we compare the standardized indicators for lung cancer morbidity in the cities in question with the urban population of foreign countries, we find the very high indicators among men in a number of cities of the USSR very interesting—they are among the highest in the world. The lung cancer morbidity indicators for women are significantly below the highest world values, and in general they are comparable with foreign indicators.

Analysis of lung cancer morbidity in the population of the cities in question, grouped in relation to industrial sectors, showed that it is highest in the group of cities possessing enterprises of ferrous or nonferrous metallurgy and chemical industry in their industrial structure, and lowest of all in the group of cities specializing in machine building and in light and food industry.

The coefficient of correlation between intensive lung cancer morbidity indicators for the male and female population in 26 cities of the USSR was equal to 0.77 ( $p = 0.0001$ ). Such a high coefficient permits the assumption that common etiological lung cancer factors exist for both sexes.

Data from the Hydrometeorological Service for 1975 were used to assess the effect of air pollution on lung cancer morbidity in the urban population. Unfortunately it is impossible to determine the relationship between lung cancer morbidity and air pollution by carcinogens, since the first information of the Hydrometeorological Service on pollution of the air basins of most of the cities in question by carcinogens—by benz(a)pyrene—is not available until 1983.

Atmospheric ecological stress is defined as the acuity of the ecological situation in connection with air pollution. It is a function of two variables—the scale of the effect produced by productive forces, and the natural environment's capacity for self-purification—that is, the atmosphere's resistance to pollution. Atmospheric ecological stress will decrease due to the environment's high capacity for self-purification, and worsen when the rate of dispersal and decomposition of pollutants is minimal. The cities included in this study are in six types of regions with different capacities of the natural environment for self-purification: from minimum in Norilsk to almost maximum in Poltava, Donetsk and Vilnius. The effect of industrial centers on the state of the air basin depends both on the structure of industrial production and on its scale.

To determine the effect of cities that are large industrial centers on the state of ambient air, a number of procedures by which to assess the effect of industrial centers on parameters of the air environment were tested out for the first time in research of this sort. The need for using several procedures is brought about by the absence of a universal method of determining atmospheric ecological stress, by the lack of information on pollution of the air basin in earlier years, and by the conditionality of existing assessment procedures.

The point assessment of the effect of industrial centers on the atmosphere (the first procedure) is determined as the result of the product of the number of points awarded for the size of the city (cities are ranked on the basis of population size) and the total number of points awarded for the structure of its industrial production [2]. Depending on the degree of its effect on the environment, each industrial sector is awarded a certain number of points, with regard for the discharges of production operations releasing cancer-causing compounds. Analysis of the results after adjusting the ranks with regard for the natural environment's capacity for self-purification made it possible to distinguish cities with a major effect on the atmosphere (Chelyabinsk, Sverdlovsk, Ufa, Donetsk, Novosibirsk) from those with a minor effect (Kursk, Poltava, Ulyanovsk, Vilnius, Voronezh).

The second procedure for assessing the effect of cities on the condition of ambient air entails calculating the quantity of harmful substances (in tons per day) released by industrial enterprises, by boiler plants and by motor transportation, per square kilometer of city area. This procedure also indirectly reflects the carcinogenic

danger, inasmuch as industrial sectors such as ferrous and nonferrous metallurgy, by-product coke industry, oil refining and petrochemistry, which are characterized by the greatest contribution to air pollution by carcinogenic substances, also produce the largest discharges. Depending on the quantity of toxic substances entering the air basin of the cities in question per square kilometer of their territory, all cities were divided into seven groups in order to determine the rank of the industrial center's effect upon ambient air. After correcting for the atmosphere's resistance to pollution, the most acute situation was revealed in Norilsk, Magnitogorsk, Chelyabinsk, Krivoy Rog, Lipetsk, Omsk and Ufa, and the least acute was revealed in Poltava, Vilnius, Voronezh and Kursk.

The third procedure by which services of the USSR State Committee for Hydrometeorology assesses the effect of industrial centers on ambient air entails determining the amount by which toxic substances polluting the atmosphere exceed the MPC [maximum permissible concentration] [1], using the formula:

$$API = \frac{1}{n} \sum_{i=1}^n \frac{q_{ave-i}}{MPC_i}$$

where API—air pollution index;  $q_{ave-i}$ —average concentration of one of the impurities,  $mg/m^3$ ;  $MPC_i$ —its average daily MPC,  $mg/m^3$ ;  $n$ —number of impurities.

A flaw in this procedure was revealed in the course of calculations using it: The more complete the pollution

data are and the larger the number of ingredients are accounted for and measured, the lower the air pollution index. The results of this procedure are not included in subsequent calculations.

Because the first and second procedures make it possible to characterize the effect of industrial centers on the condition of the air environment in relation to different supplementary parameters, an average rank for atmospheric ecological stress was derived. We found, as a result, that stress is highest in cities with developed ferrous and nonferrous metallurgy, by-product coke industry, and oil refining and petrochemical industry (Chelyabinsk, Magnitogorsk, Norilsk, Lipetsk, Krivoy Rog, Ufa, Baku), and it was the least in cities specializing in machine building and in light and food industry (Poltava, Vilnius, Voronezh, Kursk, Ulyanovsk).

The table below gives the coefficients of correlation between different lung cancer morbidity indicators for the populations of the cities in question, the atmospheric ecological stress levels and pollution of ambient air by dust, sulfur dioxide ( $SO_2$ ), carbon monoxide (CO) and nitrogen dioxide ( $NO_2$ ). It is evident from the table that a close correlation exists between atmospheric ecological stress and morbidity of the urban male population ( $r = 0.64$ ,  $p = 0.001$ ). Lung cancer morbidity among men also correlates significantly with pollution of the atmosphere by dust ( $r = 0.57$ ,  $p = 0.005$ ),  $SO_2$  ( $r = 0.67$ ,  $p = 0.0008$ ) and CO ( $r = 0.42$ ,  $p = 0.04$ ); the correlation with  $NO_2$  is weaker. The correlation between lung cancer morbidity among women and air pollution is weaker, and it is statistically insignificant ( $r = 0.38$ ,  $p = 0.06$ ).

**Coefficients of Correlation Between Atmospheric Ecological Stress and Lung Cancer Morbidity in Populations of 26 USSR Cities**

Morbidity Indicator	Sex	AES	Pollutant Ingredients			
			Dust	$SO_2$	CO	$NO_2$
Intensive	M	0.33	0.35	0.35	0.27	0.27
		(0.097)	(0.08)	(0.07)	(0.17)	(0.16)
	F	0.36	0.28	0.18	0.18	0.16
		(0.07)	(0.16)	(0.37)	(0.38)	(0.43)
Standardized	M	0.64	0.57	0.67	0.42	0.31
		(0.001)	(0.005)	(0.0008)	(0.04)	(0.12)
	F	0.38	0.34	0.11	0.28	0.10
		(0.06)	(0.09)	(0.57)	(0.16)	(0.61)
Truncated	M	0.58	0.58	0.62	0.38	0.32
		(0.004)	(0.004)	(0.02)	(0.06)	(0.11)
	F	0.34	0.37	0.13	0.25	0.13
		(0.09)	(0.06)	(0.51)	(0.21)	(0.50)

Note. The value of  $p$  is given in parentheses.

The sex-age structure of the population of Norilsk differs sharply from the structure of other cities in the study. It is dominated by men, and the age group over 50 years makes up only 6 percent of the population, which

reflected upon morbidity indicators per 100,000 population, especially among men: The ordinary indicator (21.7 percent) was the least, while the standardized (110.6) was the maximum of 26 cities. To avoid the

effect of these contrasting indicators on the correlation coefficient, we repeated the correlation analysis of the cities in question without Norilsk. The resulting correlation coefficients differed insignificantly from those calculated earlier: The coefficient between atmospheric ecological stress and morbidity of the male population ( $r = 0.59$ ,  $p = 0.004$ ) and morbidity among women now correlated significantly with atmospheric ecological stress ( $r = 0.4$ ,  $p = 0.048$ ), although the magnitude of the coefficient remained just as small.

As was noted earlier, the purpose of this study was also to determine the effect of smoking on lung cancer morbidity of the urban population. Unfortunately there are no data by which we could determine how widespread smoking is in the population. Use of tobacco articles by the population can be assessed only on the basis of sales information. It was impossible to collect exhaustive information on the sale of tobacco articles for 1970, 1975, 1979 and 1983 in quantitative and monetary terms, broken down in relation to different types of articles and in relation to all cities in question, because either such statistics were not compiled in previous years, or they were discarded. Calculations were therefore made for 12 cities for which we were able to obtain the materials of interest to us.

Per-capita consumption of tobacco articles in the adult population (over 15 years) was calculated. Sales of tobacco articles in monetary terms do not reflect the true pattern of smoking, inasmuch as there are significant differences in their consumption in different cities. In capital cities for example (Yerevan, Baku, Vilnius), consumption of filter cigarettes, which cost more than nonfilter cigarettes and "roll-your-own" cigarettes, dominates, at the same time that in Krivoy Rog, Magnitogorsk, Omsk, Ust-Kamenogorsk and Leninogorsk consumption of nonfilter cigarettes and "roll-your-own" cigarettes is higher.

The coefficient of correlation between consumption of tobacco articles per capita in quantitative terms and lung cancer morbidity among men was equal to 0.72 ( $p = 0.02$ ), while among women it was 0.73 ( $p = 0.01$ ).

Thus the results of the research permit the hypothesis that a single etiological lung cancer factor exists for both sexes—smoking. The role of air pollution in the genesis of lung cancer is significantly smaller, as is indicated by the weak correlation of this parameter with lung cancer morbidity among women. On the other hand the pronounced and statistically significant correlation between pollution of the air basin and lung cancer morbidity in the male population alone may be explained both by their exposure to carcinogens on the job and by the higher frequency of smoking.

It should be noted however that quantitative and qualitative assessment of the effect of air pollution is extremely difficult because it is impossible to isolate the independent action of air pollution from the complex of known factors owing to a lack of information or even the

absence of a retrospective quantitative assessment of air pollution, as well as because of the impossibility of determining individual exposure, especially in the past. This indicates the need for constant ecological monitoring of the state of the atmosphere and epidemiological research aimed at studying the risk of lung cancer associated with air pollution, primarily by carcinogens. We need new dose-effect quantitative epidemiological data reflecting the carcinogenic effect of individual substances polluting the air on man, data which can be used to come up with adequate maximum permissible concentrations based on data from epidemiological research.

When assessing the oncological risk associated with air pollution we need to consider smoking and occupational factors. On the other hand isolated study of the effect of air pollution on the incidence of malignant tumors without regard for known etiological factors can lead to unjustified conclusions. Prevention of lung cancer must be comprehensive, and it must include both measures against smoking and measures to protect ambient air from pollution by industrial and transportation wastes.

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**Hygienic Principles of AIDS Prevention: A Survey**  
917C0309B Moscow GIGIYENA I SANITARIYA in  
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[Article by I. E. Lalayants and L. S. Milovanova, Scientific Research Institute of Neurosurgery imeni N. N. Burdenko, USSR Academy of Medical Sciences, Moscow]

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[Text] The tragedy in Elista, and then in other cities where children were infected with AIDS in the hospital, once again attracted attention to the problem of this viral disease [1,4,16,20], which has already been illuminated rather extensively in Soviet scientific and popular scientific literature [8-15,18]. An investigation of the circumstances showed that infection occurred as a result of using unsterilized syringes. The source of the disease itself was the parents of a child, one of whom had worked earlier in Africa. The commission of the RSFSR Ministry of Health that conducted the investigation pointed to the poor knowledge of the hygienic principles of AIDS prevention possessed by physicians and nurses. It is to these principles that the authors have devoted this paper.

The "official" beginning of AIDS, or the acquired immune deficiency syndrome, goes back to 1980, when in San Francisco, and then in New York, young homosexual men who had not been ill previously were discovered to have pneumonia elicited by a normally harmless microorganism, and Kaposi's sarcoma—a rare form of tumor of vascular origin usually diagnosed in elderly natives of countries in the Mediterranean basin [8-15,18]. This form of sarcoma was described in 1872 by the Hungarian pathologist M. Kaposi in men of North Africa. Other symptoms were then discovered among these young people as well—diarrhea, herpes and fungal afflictions. Dementia, or degenerative affliction of the central nervous system, was included in the syndrome on 1 September 1987.

It was also revealed that this combination of symptoms, or syndrome, is the product of acquired immune deficiency, elicited by reduction of the number of special immune lymphocytes that have come to be called T-helpers (from the English word "help"), inasmuch as they help to "start" the immune response. As we know, the latter begins with absorption of the foreign antigen (AG) by Metchnikoff's macrophage (MP), which is an antigen to a T-helper. By means of a special protein interleukine-2 (IL-2), a T-helper in turn causes B-lymphocytes to synthesize protein antibodies and stimulates killers (from the English word "kill"), the function of which is to destroy cancer cells generated in the body and reject foreign organs and tissues. The structure of the immune response may be diagrammed as follows: Inasmuch as the T-helper is the first thing to be attacked by AIDS, it becomes understandable why the human body becomes defenseless against bacterial, fungal and viral infections. Narcotics used by homosexuals that could be toxic to T-helpers were suspected at first. However, in 1982, researchers began tending toward the idea that AIDS is viral in origin. The virus was soon discovered.

R. Gallo, who isolated human immune-deficiency virus (HIV) jointly with L. Montanye [transliterations], wrote: "The AIDS report shook the minds of physicians at the moment when the leading authorities of world medicine were predicting the imminent end of all epidemics" [18].

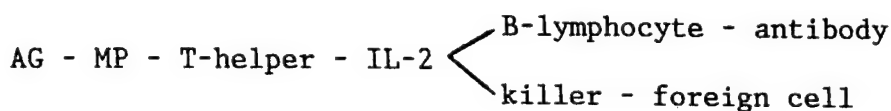
HIV is one of the tiniest RNA-containing viruses known today. Its genome contains not more than a dozen genes, one of which is the gene for its outer protein envelope, p120 "env". It is by means of this protein that the virus binds with the surface of the target cells of its attack, and T-helpers in particular. Binding of p120 protein occurs with a T4 protein receptor (CD4 in another classification), located in the cell membrane.

Protein p120 is highly variable, which is why antibodies against it are often found to be ineffective. Envelope proteins of different HIV strains may differ in their amino acid composition [44]. At the same time individual segments of the protein chain are rather stable. Thus, substitution of the amino acid tryptophan in the 432nd position on p120 protein results in loss of viral virulence and of the capacity for infecting cells. On the other hand substitution of isoleucine at position 425 does not affect virulence but does influence the ability of HIV to bind with T4. It may seem paradoxical today that antibodies may even increase the infectiousness of the virus [32].

Science has already been familiar for a rather long time with immune deficiencies, among which we can include allergies, cancer, anaphylactic shock and psoriasis. They are all genetic in nature, as are congenital immune deficiencies that arise, for example, as a result of a defect in the gene for adenosine deaminase, one of the most important enzymes of immune cells. The most famous example of this sort was a boy by the name of David in the American city of Houston, who was forced to live in a sterile tent for over 13 years, after which he was given a bone marrow transplant from his sister. Unfortunately the marrow cells became malignant, and David died.

A unique feature of immune deficiency acquired as a result of infection by HIV is the extremely long incubation period, lasting an average of 5.5 years in men and 8.5 in women, which is possibly evidence of the greater stability of female immune status. The incubation period may be reduced when an individual is infected with various forms of disease.

In fact, as was revealed in 1988-1989, the incubation period may last even longer. This was shown by means of a so-called polymerase chain reaction (PCR), which permits us to determine not antibodies in the blood of the infected person but a copy of the DNA (cDNA) of the



HIV genome in target cells [10-12,71,75]. It was shown by the PCR that viral cDNA may remain in a "silent" state in cells for a minimum of three years without manifesting itself in any way; protein antiviral antibodies would be absent from blood in this case. This makes it understandable why people may be infected by blood tested for the presence of antibodies and transfused during surgery. The PCR also clearly shows that not all children born of mothers with AIDS are infected with HIV. At the same time it was established by this reaction that the proportion of lymphocytes in peripheral blood carrying viral cDNA in their genome is significantly higher than was supposed earlier, attaining  $10^{-3}$ —that is, one lymphocyte of every thousand is infected [70].

It is presently difficult to study many aspects of AIDS in connection with the absence of a dependable model of this disease in animals. Only recently was it possible to infect some animals—mice, rabbits and monkeys in particular [27,49,64-66]. A transgenic mouse carrying an HIV gene in its genome has been obtained. When one of the HIV regulatory genes is transferred to an egg cell of such a mouse, its young are born with numerous soft tissue tumors, which emphasizes the relationship between AIDS and cancer [8].

Especially interesting data were obtained with monkeys. Calculations show that HIV may have arisen out of monkey virus 30-40 years ago (150-200 years ago according to other estimates). It is noteworthy that immune deficiency viruses isolated from African monkeys do not affect their hosts; however, they do elicit a state close to AIDS in Asian rhesus monkeys. It was believed earlier that although chimpanzees do get infected by HIV, they do not suffer AIDS. However according to the latest data, antibodies begin to appear after three years in them as well, indicating progressing illness [64]. This permits an analogy with data obtained for man by the PCR.

Animal models made it possible to answer some as yet unanswered questions about AIDS. It was mentioned above that cells are infected by the virus after its envelope protein p120 comes in contact with a T4 receptor. In recent times, however, data were obtained indicating that cells may be infected by the virus even in the absence of T4 protein on their surface; this is especially true of brain and muscle cells [36,76]. Moreover, increasingly greater amounts of data are being accumulated on the relationship of AIDS to autoimmune processes [69,77] and defective viruses eliciting diseases of other body organs and systems, particularly the thyroid gland, and leukemia [55,74]. Nothing more definite can be said about this at this time because of the absence of experimental results that could be dependably reproduced in animals.

In the therapeutic aspect, AIDS still remains an incurable viral disease. Azidothymidine (AZT), which is available under various commercial names (Zidovudin, Retrovir [transliterations] etc.), has recommended itself

reasonably well as a therapeutic agent. The advice since quite recently is to prescribe it not only to patients who had experienced a sharp decline in the number of T-helpers but also to infected individuals with a constant quantity of immune cells [29,43,50,66]. Permission has also been granted for using AZT on children, which sharply widens the group of people employing this drug, one which disturbs the action of the viral enzyme synthesizing cDNA on a viral RNA matrix at the molecular level.

Unfortunately, an HIV strain resistant to AZT was isolated in 1989 [29,58,62]. This is why we need to resume the search for effective drugs. We also need to consider the fact that AZT is not a medicine against AIDS, inasmuch as it can only retard development of infection in the body, while it is unable to destroy it, and it cannot affect viral cDNA in the cell genome. This is why researchers have shown interest in a report of isolation of a second viral enzyme responsible for splitting of proteins of HIV in the course of its replication and duplication [31]. Knowledge of the molecular mechanisms of the action of this enzyme may help in the development of effective methods of treating this viral infection.

For the moment scientists are trying to proceed along the path of creating bioengineering resources, particularly so-called immunoadhesins, which are soluble forms of cellular T4 protein that bind with the surface of viral particles and block their penetration into the cell [12,34]. In addition, molecules of cytotoxic substances—for example a toxic antibiotic or the plant poison ricin, which bind with viral p120 protein on the surface of infected cells and kill them—"fasten themselves" to molecules of immunoadhesins.

Various vaccines are also being developed out of cowpox and poliomyelitis virus, the genome of which is introduced by means of bioengineering methods into the HIV gene. Experimental vaccines are being tested on people and animals. The first results of testing a vaccine made from killed HIV were obtained with chimpanzees [44,56,66]. Bioengineering developments are illuminated in greater detail in other surveys [8,10-12,15].

AIDS has become not only a biomedical but also a political problem in the modern world [30,33]. The U.S. Congress allocates around \$1.5 billion yearly to the fight against this menacing disease. At the same time politicians condoned the spread of homosexuality and drug addiction for a rather long time, fearing to trample the interests of certain circles, and thus promoting the spread of HIV. Now that more effective expenditure of public assets in the fight against AIDS is becoming important, the USA's social welfare agencies realize that simply frightening the people won't help, inasmuch as they quickly find some other means to protect themselves from such fears [33].

More promising in this regard is the Global AIDS Control Program, developed by the World Health Organization (WHO) under the guidance of Dzsh. Mann [transliteration] [54]. The U.S. Academy of Sciences has also published its recommendations [61]. They essentially call for prevention, and particularly for safe sex using condoms, inasmuch as AIDS is a sexually transmitted disease. Introduction of safe sex led to a decrease in the number of new cases among San Francisco's homosexuals.

Compulsory mass testing, which foreign experience has shown to be expensive and rather ineffective, is a complex issue. Nonetheless, some countries, for example Bulgaria and Cuba, are conducting such tests [42,48]. Cuba is also isolating all revealed infected individuals and patients in a sanatorium set up in a hacienda—a former possession of Fink los Kosos [transliteration] 15 km from Havana. In 1989 it housed 264 infected individuals, including 73 women and two hemophiliacs who received HIV with transfused clotting factor [57].

AIDS prevention presupposes a knowledge of the paths of its spread through particular risk groups—primarily homosexuals, prostitutes and drug addicts. As we know, all three groups quite often overlap. Considering the large size of these three groups in the modern world, especially in cities, and their huge numbers of daily contacts, it is not difficult to see that even disseminating AIDS information is not an easy task in this environment [26,66]. Even among American clergy there are up to 20 percent homosexuals [67].

These risk groups can be classified as "active." At the same time there are "passive" risk groups—children, medical workers, recipients of blood and other biological tissues, sperm in particular, hemophiliacs, women in childbirth and so on [9,38,66,72,73]. In two years (1987-1989) the number of sick and infected children in the USA doubled, attaining 1,000 [47]. At the same time drug addicts also suffer a passive threat, inasmuch as together with heroin they introduce toxic metabolites into their bodies—particularly aflatoxin B<sub>1</sub> from the mushroom *Aspergillus flavus*, which kills immunocompetent cells [50]. The frequency of infection by KhTLV-1 [not further identified] virus, which elicits T-cell leukemia, has also increased among drug addicts [59].

It is interesting that circumcision, which is practiced in some African countries, reduces the risk of AIDS infection by a factor of 5-8 [70]. This once again brings attention to the various modes of infection of representatives of different races. As we know, white men do not undergo circumcision, and still, the risk of infection does not increase among them in this connection.

Thus a survey of 1.7 million American army servicemen conducted by the Pentagon revealed 2,232 infected individuals, which corresponds to a frequency of 1.3 per thousand [40]. The highest frequency (2.1 per thousand) was noted in the 25-29 year age group—that is, in the

sexually most active ages. In this case the rate of infection among whites was only 0.8 per thousand, while among blacks it was 2.9 per thousand. The next closest frequency of infection was among Hispanics (2 per thousand). The same ratio was noted earlier in the civilian population as well. We can add to this that enlisted and unmarried men were infected twice more frequently than married officers and servicemen. These data show that improper relationships promote the spread of AIDS [63].

A survey of up to 30 percent of newborn infants in 30 states is presently going on in the USA [46]. Such a survey conducted in 1986 in Massachusetts revealed two infected individuals per thousand, while in New York the figure was 8.6 per thousand statewide in 1987. The rate of infection in New York itself attained 16.4 per thousand. This is a very high indicator, which is why the question as to the nature of the revealed antibodies immediately arose: Do they belong to the mother or the child?

We should turn attention here to the nature of the first-generation diagnosticums themselves, which determine not the virus itself but AIDS antibodies. This reaction is called ELISA (from the English for "Enzyme Linked Immune Sorbent Assay"), or REMA—"Enzyme-Labeled Antibody Reaction." The latter reveals presence of antibodies in blood that are unable to neutralize the virus for some reason. This permits the notion that the REMA detects antibodies having a nature that has not yet been fully clarified. This is especially true in relation to children.

Contemporary data show that AIDS arose long before its "official" beginning—there was simply no way to detect it then [18,53]. R. Gallo noted that the AIDS outbreak marked the end of the "permissive society," with its emphasis on promiscuity [41] and drug use [37,46]. Social conditions under which even infected prostitutes have to "work" in the streets also make a great contribution to the spread of AIDS [51,52,60,68]. Tests conducted in Glasgow showed that the proportion of infected prostitutes is 75 percent here, while a 1.5-time decrease in the number of T-helpers in seropositive prostitutes was noted in Senegal's capital Dakar, which raises the risk of heterosexual transmission of HIV. It should be considered that heterosexual infection can occur as a result of a single act of intercourse [28] with an infected partner. This pertains especially to clients who do not wish to use condoms. Thus in summer 1988, at a conference in Stockholm devoted to AIDS research, a documentary film was shown about young prostitutes in Rio de Janeiro who are paid three times the price by their clients if the former agree to have sex without a condom [39].

The channels of information pertaining to AIDS have now been opened extremely wide [24]; enormous sums of money are being allocated for this purpose. Each year the USA allocates up to \$1.5 billion to its AIDS control

program, inasmuch as it feels that "economy of prevention" would save billions in the future [46]. U.S. Surgeon General E. Koop made an appeal for responsible behavior in the AIDS era, while at the same time admitting that the government is not doing enough to prevent the spread of HIV among drug addicts. There are now 1.2 million drug addicts in this country who inject drugs intravenously, and only 150,000 of them are in treatment [45].

And what is being done in this aspect in our country? Despite the availability of information and the legislative protections afforded to public health [1-5,7,17], the AIDS problem remains complex and unresolved. Its aspects in our country are somewhat different from Western countries, which are not experiencing a shortage of disposable syringes and condoms, and where AIDS is nonetheless a serious threat to public health. "There is no panic," USSR Academy of Medical Sciences President V. I. Pokrovskiy declared not that long ago in one of his interviews [21-22]. At the same time it is pointed out that our first patient was revealed in summer 1987, and as of the end of 1989 we had as many as 400 infected individuals. Development of their infection could be arrested only with AZT [19]. In comparison with Western countries, we have many infected children and women. The number of the latter is equal to the number of infected men (in Western countries this ratio is 1:10), which is an indication of a high percentage of infection of prostitutes. At the same time risk groups such as homosexuals and drug addicts are "clean" in our country for the moment (they have not been tested with the PCR). Millions of donors have been tested; individual virus carriers have been revealed among them [30].

Soviet engineers proposed a new electrothermal method of determining antibodies in the blood of subjects that makes testing easier, cheaper and faster [6]. Moscow's Infection Hospital No 2 was allocated 15 million rubles in the current five-year plan for construction of new buildings for AIDS patients [2]. An agreement has been reached with foreign companies to build the first plants producing disposable syringes [17]. The first books and pamphlets devoted to the AIDS problem have been published [15,23,25].

Truthful and scientifically substantiated AIDS information is extremely necessary, inasmuch as the opinion still exists that HIV can be transmitted with saliva when kissing, in public restrooms, by a handshake, through sports activities and by a mosquito bite [7,15,35]. While all of these myths have long been refuted, they are still being perpetually "reanimated," sowing panic and fear in the population. It is asserted for example that athletes have been placed in the risk group because they have "many male and female fans." But this is typical not only of sports but also of the young, sexually active population in general [7]. It was shown experimentally that mosquitos cannot serve as HIV carriers simply in view of their biology—another thing that has long been known [35].

We already noted that fear is a poor adviser and helper in the fight against such a menacing disease as AIDS. This is why it would hardly be worth accepting it as our ally. Quite the reverse: Constant and consistent information provided to the broadest strata of the population and specialists should be seen as the most suitable and useful way to go.

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### Evaluation of AIDS Knowledge Among Schoolchildren

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[Article by I. V. Kuznetsov and T. I. Kuznetsova, Palace of Public Health Education, Novokuybyshev]

UDC 616.98:578:828.6]:[614.374

[Text] The number of AIDS cases in the world is increasing with every year. The number of carriers of human immune deficiency virus (HIV) and of AIDS patients is growing in our country.

Given specific defenselessness against AIDS, the personal factor—that is, the moral behavior of the population and raising the occupational responsibility of medical workers—is acquiring primary significance in preventing this disease.

Since the moment that the first AIDS cases appeared in our country the Palace of Public Health Education organized extensive public health education work in the population. Special attention was turned in this case to secondary school, state vocational-technical school and [tekhnikum] students and to young workers of the city's enterprises and institutions.

One hundred questionnaires bearing 13 questions were distributed among 8th-10th year students of two schools in order to clarify what is known about AIDS. Eighty-five of the questionnaires were collected and processed.

Thirty-four percent of the respondents obtained AIDS information from the journals ZDOROVYE, SMENA and YUNOST, 15 percent did so from newspapers, 12 percent from popular scientific literature, 24 percent from discussions and lectures delivered by medical workers and educators, 10 percent from television and films, and 5 percent of the respondents from other sources.

When asked what the definition of AIDS is, only 17 (20 percent) of the individuals noted that it is a disease involving a decrease in the body's protective functions—that is, immune deficiency. All other students were unable to define the word.

Forty-nine (57.6 percent) of the individuals were unaware of the disease's origin, 25 (29.4 percent) stated that it had been imported from the African continent, three (3.5 percent) responded that the AIDS agent (HIV) was created in the laboratory, and eight (9.4 percent) did not answer the question.

A virus was named as the disease agent by 53 (62.4 percent) students; 32 (37.6 percent) had no idea what the agent of AIDS is. The pathways by which the virus is released from the body into the environment were named by 57 (67 percent), including with blood—36.3

percent, with the products of sex organs—17.4 percent, and with mother's milk—13.3 percent.

Of the questionnaire respondents, 53.6 percent did not know how stable the virus was and how long it survived in the environment, while 28.8 percent answered that the virus lives outside the human body for a long time and 17.6 percent answered that it lives for an unknown period of time.

Eleven (12.2 percent) of the respondents did not know the clinical manifestations of AIDS, while the other 74 persons named 131 signs of AIDS.

When asked questions about the length of illness and its outcome, one out of every two (49.5 percent) stated that the disease is long-lasting and incurable. The rest did not answer the question; students apparently understate the dramatic consequences of AIDS.

When asked how widespread AIDS is in our country, 45 (52.9 percent) of the individuals cited figures reflecting the situation at the moment of the survey, 21 (24.8 percent) of the students did not have such information, 13 (15.3 percent) stated that there was a large number of patients and virus carriers, and six (7 percent) said that this number was insignificant.

Questions on the pathways of AIDS transmission received 147 answers: Infection by the sexual pathway was indicated by 47 (33.3 percent) persons, by blood during transfusions—39 (23.4 percent), as a result of injections by nonsterile syringes—21 (14.8 percent), during intrauterine development and birth of a child—13 (9.2 percent), through mother's milk—five (3.5 percent), through saliva when kissing—four (3 percent), and 18 (12.8 percent) of the respondents gave no answers.

One question in the survey asked who was threatened by AIDS most often. It received 141 answers. Most respondents—125 (88.6 percent) named persons leading an amoral way of life, including 40 (28.3 percent)—prostitutes, 38 (26.9 percent)—homosexuals, 47 (33.4 percent)—drug addicts, six (4.2 percent)—blood recipients, four (2.8 percent)—children of sick mothers and six (4.2 percent)—persons applying to medical institutions.

When asked about the ways to fight AIDS, 55 students offered 127 proposals, including the following: prompt and complete revelation of AIDS patients and virus carriers (41.7 percent), introduction and wide use of disposable systems and instruments for medical procedures (18.9 percent), increasing the production and sale of condoms (12.6 percent), intensifying control over sterilization of instruments used in procedures by therapeutic and preventive institutions (8.7 percent), raising personal responsibility of people (7.9 percent), widening and improving educational work in the population (5.5 percent), and enacting stiffer measures against drug addicts and prostitutes (4.7 percent).

Our research showed that students have an insufficiently complete idea about AIDS, the features of its manifestation, the pathways of infection and prevention measures. A significant part of the respondents underestimate the negative role of immoral behavior in the spread of AIDS. Proposals on controlling AIDS made references to the need for applying extremely drastic measures against AIDS patients and sources.

In order to increase and deepen the AIDS knowledge of students, we need to conduct scientifically supported educational work, emphasizing the need for maintaining a humanitarian attitude towards AIDS patients and HIV carriers. ©COPYRIGHT: Izdatelstvo "Meditsina", 1990

### Gas-Chromatographic Analysis of Organophosphorus Insecticides in Tobacco Smoke

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[Article by F. G. Zhitku, Moldavian Scientific Research Institute of Tobacco, Kishinev]

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[Text] Wide application and frequently uncontrolled use of pesticides led to serious negative consequences in recent decades despite the fact that high hygienic requirements are imposed on the toxic chemicals employed. They include preventing their accumulation in environmental objects, their negative influence upon living organisms, acute intoxications of man and animals, unfavorable remote consequences and so on [3].

The problem of residual quantities of pesticides in raw materials, and all the more so in smoke, has important hygienic significance. Attention to pesticide residues in tobacco is associated with the possibility of their transfer to tobacco smoke and greater harm to the smoker's health.

The need for studying this problem stems from the fact that although considerable data had been accumulated and published on the dangers of smoking [6,9], tobacco consumption increased by 75 percent in the last 20 years, and will increase by another 50 percent by the year 2000 [5].

The USSR Ministry of Health is currently taking steps to reduce the harmful effects of pesticide residues. Thus in GOST [All-Union State Standard] 8073-77 and GOST 8072-77 on tobacco leaf, Section 2 of both standards was amended: Residual quantities of pesticides in tobacco leaf must not exceed the maximum permissible level (Decree No 1137 of the USSR State Committee for Standards, 2 April 1987).

But this does not actually solve the problem. Compliance with the GOST requires the corresponding methods of

determining and maintaining systematic toxicological control over pesticide residues in tobacco.

With this in mind, the Moldavian Scientific Research Institute of Tobacco has been conducting scientific research since 1981 with the purpose of creating and developing methods of determining pesticide residues in tobacco and tobacco smoke. Using the highly sensitive and selective method of gas-liquid chromatography (GLC), we have developed a procedure for determining both specific compounds (pirimor [1], geterofos and basudin [2], and selekron and volaton) and multi-ingredient pesticide mixtures.

This paper describes a GLC method permitting determination of six organophosphorus insecticides (OPI's) simultaneously in one tobacco condensate sample: basudin, geterofos, carbofos, metaphos, rogor and etafos. Special attention was devoted to OPI's because these toxic chemicals are the most toxic and harmful to human health. Data on transfer of some OPI's into the main stream of tobacco smoke available in the literature [4,7,8] are extremely meager, which predetermined the goal of our work.

### Materials and Research Methods

A Tsvet-106 chromatograph with a thermionic detector was used in the work. Chromatographically clean OPI standards were obtained from the All-Union Scientific Research Institute of Chemical Plant Protection Resources. Working concentrations of standard solutions (in acetone) of basudin and geterofos were 0.5, 1 and 5 ng/μl, carbofos, metaphos and rogor—1, 2 and 10 ng/μl, and etafos—2, 4 and 20 ng/μl.

The compounds were divided in a 200 cm x 3 mm column containing 3 percent OU-17 on N-super chromatone (0.16-0.20 mm). The chromatographic conditions were: column temperature 180°C, evaporator temperature 210°C; velocity of carrier gas (especially-pure nitrogen) 60 ml/min, of hydrogen 17-18 ml/min, of air 200 ml/min; electrometer scale  $100 \times 10^{-12}$  A, recording tape speed 200 mm/hr.

Under these conditions the relative time of retention (in relation to metaphos) was 0.45 for basudin, 0.52 for geterofos, 0.81 for rogor, 1.0 for metaphos, 1.19 for carbofos and 1.50 for etafos.

The linear range of detection was 0.5-5 ng for basudin and geterofos, 1-15 ng for carbofos, metaphos and rogor, and 2-30 ng for etafos.

The average predetermined quantities of OPI in tobacco condensate were: basudin—92.4 +/- 3.27 percent, geterofos—86.8 +/- 3.12 percent, carbofos—96.1 +/- 4.49 percent, metaphos—91.2 +/- 3.28 percent, rogor—95.4 +/- 3.46 percent, etafos—84.8 +/- 4.72 percent (the number of parallel determinations was five,  $\alpha = 0.95$ ).

A 200 cm x 3 mm column containing 5 percent SE-30 on N-AW DMCS chromatone (0.25-0.315 mm) was used as an alternate column.

Model experiments were run with instruments made by the Kauni [transliteration] Company (FRG) in order to study the extent of transfer of OPI's to the main stream of tobacco smoke. Cigarettes containing precisely predetermined quantities of insecticides were smoked using an RM/20 CS smoking machine under standard, universally accepted conditions: draw volume 35 ml, draw time 2 sec, interval 58 sec. Condensate from the smoke of 10 cigarettes (from the solid-liquid phase of the main stream) was trapped in an electrostatic trap at 15-17 kV. The gas phase (the main stream) of the smoke was bubbled through 100 ml acetone in a Drechsel flask.

In order to ensure reproducibility of the results, cigarettes were selected by weight (+/- 10 mg) and draw resistance (+/- 5 mm Hg), and brought to standard humidity (13 +/- 1 percent).

One of the most important tasks that had to be carried out was to verify Guthrie's assertion [7] that in model experiments, predetermined quantities of pesticides could be introduced directly into cigarettes with a microsyringe, or cigarettes could be made with tobacco to which predetermined quantities of the compounds had been applied. The way the quality (class) of the tobacco affects the extent of transfer of OPI's to the main stream of tobacco smoke also had to be established using cigarettes made by the Kishinev Tobacco Combine—Kosmos, Doyna and Astra (nonfilter). The contribution of two different filters—Soviet and ones made by Austria's Privileg (both are acetate filters)—to reducing the concentration of OPI residues in tobacco condensate needed to be evaluated.

The coefficient of variation was used as the criterion of reproducibility of the results of the analysis. The standard deviation, the relative standard deviation and the confidence interval of the mean were calculated using Student's  $T_{\alpha,k}$  significance test with  $n = 5$  and  $\alpha = 0.95$ .

The chromatograms were subjected to quantitative treatment using the method of relating peak height to the standards. The concentration of pesticides in an analyzed sample was calculated as the mean of three parallel determinations. Calculations were carried out using the commonly accepted formula [2].

#### Procedure for Determining OPI's in Tobacco Condensate (Developed by the Author)

Tobacco smoke condensate collected in the electrostatic trap is washed off with acetone (5x5 ml). Acetone extract is concentrated in a rotary vacuum evaporator to a volume of 5-6 ml, 10 ml chilled aqueous solution saturated with sodium chloride are added, and the extract is then placed in a refrigerator at 10-12°C for 30-40 min.

The water-acetone extract is filtered through filter paper (blue ribbon) directly into a separating funnel. The flask is washed by two more batches of 5 ml chilled 50 percent aqueous acetone, which is also transferred to the filter. Then 225 ml of 50 percent aqueous acetone are added, and all preparations are re-extracted in chloroform at a rate of 3x25 ml, with the funnel being shaken for 1 minute each time. The combined chloroform fraction is washed with 2x10 ml 0.01 N NaOH and 1x10 ml 0.01 N HCl, dried by filtration through a layer of anhydrous sodium sulfate (10 gm), and evaporated under a vacuum at 40-45°C. Solvent traces are removed in a flow of cold air. Exactly 1-5 ml acetone are added to the dry residue, and then the latter is chromatographed.

#### Results and Discussion

The experimental results are shown in tables 1 and 2 (pesticides were not detected in the gas phase of the main stream).

**Table 1. Effect of the Means of Introducing Prescribed Quantities of Compounds (OPI's) on the Degree of Their Transfer to Tobacco Condensate of Different Brands of Cigarettes**

Insecticide	Introduced, $\mu\text{g}$ (10 cigarettes)	Degree of Transfer, %			Coefficient of Variation, %			Degree of Transfer, %			Coefficient of Variation, %			Degree of Transfer, %	Coefficient of Variation, %
		A	B		A	B		A (1)	B (1)		A (1)	B (1)			
		1	1	2	1	1	2	1	1	2	1	1	2-	Astra 1	
Basudin	20	4.9	6.7	10.2	11.7	8.2	4.0	3.7-	4.9	5.7	10.9	10.3	4.5	22.5	6.2
Geterofos	20	5.9	10.7	12.9	11.1	6.6	4.9-	2.5	2.0	6.5	11.4	14.6	5.1	19.1	4.9
Rogor	200	1.2	1.5	2.7	8.6	8.0	6.4	1.1-	1.1	0.6	8.9	9.6	7.4	1.2	8.3
Meta-phos	100	1.3	1.2	2.7	11.1	13.5	4.6	0.3-	1.1	1.1	10.1	6.4	5.1	2.5	7.9
Carbofos	100	1.7	1.1	3.1	7.8	10.7	5.4	0.7-	1.6	1.9	8.1	10.8	6.0	3.9	6.6
Etafos	200	2.6	3.6	7.9	7.2	9.7	2.4	1.9	-1.7	3.4	6.9	9.5	3.2	8.5	6.8

Note: Here and in Table 2: A—mass-produced Kosmos cigarettes, B—Kosmos cigarettes with Privileg filter, A (1)—mass-produced Doyna cigarettes, B (1)—Doyna cigarettes with Privileg filter; 1—compounds introduced into cigarettes with microsyringe (method 1), 2—preliminary treatment of tobacco with compounds, followed by addition of Privileg filter to cigarette (method 2)

Table 2. Degree of Absorption of OPI's by Filters and Their Residues in Astra Cigarette Ashes

Insecticide	Introduced, $\mu\text{g}$ (10 cigarettes)	Degree of Absorption, %			Coefficient of Variation, %			Degree of Absorption, %			Coefficient of Variation, %			Residues in Ashes, %	Coefficient of Variation, %
		A			B			A (1)			B (1)				
		1	1	2	1	1	2	1	1	2	1	1	2-		
														Astra 1	
Basudin	20	5.6	10.5	12.8	9.4	9.9	3.7	9.4-	9.7	12.9	9.6	8.5	4.2	6.2	3.2
Geterofos	20	3.1	10.8	13.7	11.0	10.5	4.8	-3.7	8.3	12.7	10.1	11.9	5.0	5.1	5.0
Rogor	200	0.6	2.9	3.5	8.2	8.5	5.1	2.0	-2.9	3.1	8.4	8.8	5.9	1.1	5.9
Meta-phos	100	0.5	2.0	2.9	16.3	10.1	4.3	0.8-	3.1	4.0	11.3	6.9	4.9	0.6	10.7
Carbofos	100	0.9	4.8	7.5	10.9	9.9	5.5	1.4-	6.2	7.1	10.2	8.3	5.9	1.1	3.8
Etafos	200	1.8	5.5	6.8	13.2	4.7	3.1	2.2-	5.3	6.4	11.5	7.2	3.2	1.8	3.1

Analyzing the obtained data we conclude that when conducting experiments to study the degree of transfer of pesticides into tobacco condensate during smoking, the means by which standards are introduced has especially important, decisive significance, determining the dependability and reproducibility of the end results. Thus it is evident from Table 1 that preliminary introduction of pesticides into tobacco (method 2) provides more satisfactory results (the coefficients of variation for Kosmos and Doyna cigarettes are 2.4-6.4 and 3.2-7.4 percent respectively), as compared to introduction of the compounds with a microsyringe directly into the cigarette (coefficients of variation 7.2-11.7 percent for Kosmos cigarettes and 6.4-14.6 percent for Doyna cigarettes). This is apparently explained by differences in the distribution of the compounds in the cigarette. The fact is that when pesticides are nonuniformly distributed (method 1), during the interval—that is, when the cigarette is smoldering undisturbed, large quantities of pesticide may wind up in this zone; in response to high temperature they break down and are volatilized into the passive smoke stream or remain in the ash. This leads to large losses of pesticides, which wind up in the passive smoke stream, to a correspondingly low percentage of their transfer to condensate (for Kosmos—1.2-4.9 and 1.1-10.7 Privileg filter), for Doyna—0.3-3.7 and 1.1-4.9 (Privileg filter), and to wide scatter of the coefficients of the variation.

It should be noted that in some cases compounds can be introduced by method 1 uniformly with a microsyringe: Comparing the degree of transfer of geterofos to condensate in relation to methods 1 and 2, we can see that these values are very close, as are the corresponding coefficients of variation (6.6 and 4.9 percent). Similar results were also obtained for metaphos. However, the advantage of method 2 is practically indisputable as being methodologically more acceptable and reproducible.

Attention should be turned to the difference in the degree of transfer of OPI's to tobacco condensate depending on cigarette brand. Thus from 2.7 percent

rogor to 12.9 percent geterofos are transferred to condensate of Kosmos cigarettes (Privileg filter, introduction method 2), while the transfer rates for Doyna condensate are 0.6 percent rogor and 6.5 percent geterofos—that is, they are several times lower. In our opinion this fact may be explained by a mutual relationship between the quality of the tobacco in the cigarette and its burning quality, and consequently with the number of drags (the average number of drags is 10-11 for Kosmos cigarettes, 12-14 for Doyna and 15-17 for Astra).

Different degrees of absorption of OPI's by filters (see Table 2) are a consequence of using different methods of introducing the compounds and using filters of varying quality. The higher filtering capability of the Privileg filter is probably explained by its improved technological properties. On the other hand close values for the degree of absorption of OPI's by the Privileg filter irrespective of cigarette brand may be the result of the fact that owing to selective adsorption, the filter becomes saturated with each of these compounds, after which the OPI's pass through the filter without being arrested, and the quantity of compounds adsorbed by the filter remains unchanged. Thus the filter makes a certain contribution to reducing the quantity of pesticides transferred to tobacco condensate.

If we use data on the degree of transfer of the compounds to condensate and the quantity of compounds trapped by the filter to calculate the total losses of OPI's during smoking, we find (using Kosmos, Doyna and method 2 as an example) that they are 77.0-85.4 percent for basudin, 73.4-89.7 percent for geterofos, 93.8-96.5 percent for rogor, 94.4-96.8 percent for metaphos, 89.4-94.1 percent for carbofos and 85.3-93 percent for etafofos. In the case of Astra nonfilter cigarettes, the OPI losses are estimated within the same limits—from 77.5 to 98.8 percent.

It should be noted that our data are consistent with those found in the literature. Thus the degree of transfer of rogor and basudin into condensate is 1 and 10 percent respectively according to [4], while according to our

results it is 0.6-2.7 and 5.7-10.2 percent. According to [8] the degree of absorption of carbofos by the filter hardly differs from our result (correspondingly 8.8 and 7.1-7.5 percent), though some discrepancy does exist in the degree of transfer of the compound into condensate—7.0 and 1.9-3.1 percent.

It was established on the basis of the OPI's studied here that significant losses of compounds of this group may occur during smoking. However, inasmuch as the possibility of formation of products of greater toxicity is not excluded, correct toxicological assessment of pesticide residues in tobacco smoke is possible only with regard for all of the resulting pyrolytic products.

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### Japanese Assistance in Eliminating Chernobyl Effects

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2nd edition p 6

[Article by S. Troitskiy: "Japan—To Chernobyl"]

[Text] A trip to Chernobyl ended a visit to our country by a group of leading scientists specializing in radiation medicine from Japan.

The authoritativeness of this delegation can be assessed from just a few names. I. Sigematsu—general director of the Institute of Radiation Effects, H. Matsudaira—director of the National Institute of Radiation Medicine, A. Kuramoto—director of the Scientific Research Institute of Nuclear Medicine and Biology of Hiroshima University, T. Kumatori—director of the Association of

Radiation Effects, S. Nagataki—professor of the medical school of Nagasaki University. Representatives of the government and foreign affairs ministry of Japan were also in the delegation.

Their work began in Moscow, where talks were conducted for three days on possible ways of interacting to solve Chernobyl's problems with the leaders of the Soviet Union's State Committee to Eliminate the Consequences of the Accident at the Chernobyl Nuclear Power Plant, the corresponding committees of the RSFSR, Belorussia and the Ukraine, and with the most prominent Soviet scientists in this field. The basic directions of future cooperation were worked out. The Japanese side is prepared to provide the necessary equipment and medicines.

### Calculation of Escape of Radon Isotopes From Soil Into Atmosphere During Surveying Operations and Design of Residential Buildings

917C0391 Moscow *PROYEKTIROVANIYE I INZHENERNYE IZYSKANIYA in Russian* No 6, Dec 90 pp 17-19

[Article by Yu. N. Soroka, candidate of technical sciences, and A. I. Molchanov, engineer, Ukrainian Branch of the All-Union Scientific Research and Design Institute of Industrial Technology]

[Abstract] Engineering project surveys must include careful assessment of the construction site with measurement of the escape of radon from the ground and the power of the exposure dose of  $\gamma$ -radiation to test the suitability for residential construction in the area assessed. Residential construction should not be placed on sites on which the mean escape dose of radon isotopes is more than 70  $\mu$ R/hour and the density of radon flow is more than 80 mBeq/(m<sup>2</sup>s from the residential construction or the construction must be carried out according to special designs for especially stipulated conditions. The amount of radon escape from soils must be calculated during the design of residential buildings and appropriate steps must be taken to prevent over-irradiation of people. Highest concentration of radon escape in residential buildings must be expected in the night and the morning. Expected radon concentration on the first floor of future buildings may be calculated by the formula  $E_{Rn} = [C_{atm} + (3.6 \times 10^{-3} S F K) / (\lambda_v V)] n$ , where  $E_{Rn}$  is the effective equilibrium concentration of radon in the premises, Bk/m<sup>3</sup>;  $C_{atm}$  is the radon concentration in the free air used for ventilation, Bk/m<sup>3</sup>; S is the floor area, m<sup>2</sup>;  $\lambda_v$  is the rate of air exchange, hr<sup>-1</sup>; V is the volume of the premises, m<sup>3</sup>; F is the density of flow of radon from the soil, Bk/(m<sup>2</sup>xs<sup>-1</sup>); K is the coefficient of change of flow by the floor and foundation; n is the coefficient of equilibrium in the premises between radon and its daughter products of decay. A table showed rate of radon escape from the soil in 11 cities or regions of the USSR. Figures 1; references 3: Russian.

# **Problems of Echinococcosis and Measures to Control It**

917C0328A *Frunze ZDRAVOOKHRANENIYE KIRGIZII in Russian No 6, Nov-Dec 90 pp 3-4*

[Article by M. A. Aymanbetov, S. A. Abdyrasulov, and A. Z. Mukhamedzhanov, Biology Department of the Kirghiz State Medical Institute under the "Social Hygiene and Organization of Public Health" rubric: "Problems of Echinococcosis and Measures to Control It"]

[Text] Echinococcosis is difficult to control because the parasite possesses great fertility not only in the mature but also in the larval stage; the latter attacks about 60 kinds of herbivorous and omnivorous mammals (sheep,

goats, cows, horses, etc.) and man, and adult attacks dogs, jackals, wolves, and foxes.

I.K. Akhunbayev (1965), Yu. A. Volokh (1965), B.A. Akmatov (1988) Sh. D. Dzhumadilov (1967, 1978), and others have studied echinococcosis problems. However, there is no work completely concerned with the characteristics of the epidemiological and epizootic processes in foci of echinococcosis in Kirghizia.

We studied clinical patient and dissection data of the city of Frunze in an effort to explain the affliction of the population of the republic with echinococcosis.

For the period from 1984 to 1986, 636 patients were hospitalized because of echinococcosis (Table); 592 with a diagnosis of "echinococcosis," and 44 with "alveococcosis."

**Distribution of Patients With Echinococcosis and Alveococcosis Hospitalized in Frunze City Clinics, By Place of Residence**

Years	Chuskiy Zone		Issyk	Naryn	Talas	Osh	Dzhambay	Kochkor	Total
	Frunze	Subordinate Republic							
1984	23	28	29	9	8	12	8	8	125
1985	32	34	21	13	5	7	6	124	
1986	26	29	20	12	6	11	5	4	113
1987	31	33	28	13	7	10	7	7	136
1988	30	32	27	15	5	14	10	5	138
Total	142	156	125	62	31	54	36	30	636

Analysis shows that at present there is a moderate rise in the echinococcosis morbidity of the population, and the frequency of the identification of infected persons is increasing.

Of 142 patients in the city of Frunze, the majority comprised students coming from agricultural areas. Thirty patients (4.6 percent) came from one Kochkorskiy rayon alone, and of them, three (10 percent of all patients with echinococcosis) were stricken with alveococcosis.

Alveococcosis is encountered more frequently in the Kochkorskiy rayon than in others. Sh. R. Dzhumadilov (1979) came to the conclusion that the region is a natural focus of human alveococcosis.

According to the data of the total number of patients with echinococcosis, 270 (43.7 percent) were men, and 242 (38 percent) were women; among children, there were 124 cases (18.24 percent); 68 (54 percent) were boys and 56 (46 percent) were girls. According to the data of the majority of researchers, the echinococcosis morbidity among women is always higher than among men. According to the data of N.K. Studentsova (1948), children below ten years of age comprise 0.8 percent of all echinococcosis patients of Kirghizia; according to the data of Yu.A. Volokh (1963), 10.25 percent; and

according to the research results of B.A. Akmatov (1988), children below the age of 14 comprised 28 percent of all echinococcosis patients.

Thus, according to the data of B.A. Akmatov and our data, the echinococcosis morbidity rate of children in Kirghizia is increasing. This is explained by the fact that children have become more actively involved in livestock farming, are more frequently in contact with dogs, and do not observe the rules of personal hygiene.

According to the national composition, the highest percentage of echinococcosis patients comprise Kirghiz (551), Russian and Ukrainian, 69 (19.85 percent), Dungan, nine (1.5 percent), members of other nationalities, five cases (0.95 percent). It is well-known that the Kirghiz basically are employed in agriculture. Every year the number of agricultural farms increases, and therefore the probability of expansion of the geographic range of the helminth also increases. Kirghiz stockmen slaughter sheep out-of-doors right beside dogs, to which they throw out the internal organs (frequently with echinococcus cysts). Thus, stockmen, shepherds, and their families comprise higher risk groups. In addition, dogs are not examined to find out if they are helminth carriers, or if they are examined, it is done as a formality. Dogs can host more than 200-250 parasitic adult worms in their intestine. And each parasite in a very short time can

discharge up to 1000 eggs. They mature and develop intensively and continually. The eggs, excreted with the feces of the dog, fall on soil, grass, vegetables, water, even in milk and food. Eggs are spread by the wind to pastures, mountains, and rivers, and contaminate them. The onchosphere of the parasite is very resistant to unfavorable abiotic factors. At a low temperature, the onchosphere falls into a state of anabiosis and at the same time the viability of the echinococcus eggs is retained under the snow until the following season.

By analyzing the composition of echinococcosis patients according to social position, we came to the conclusion that 450 (72.5 percent) of the patients resided in an agricultural area.

In order to decrease echinococcosis morbidity, it is necessary to carry out the following organizational measures:

- organize a laboratory for helminthology or parasitology at the Scientific Research Institute of Ecology of the republic;
- conduct mass detailed examination of stockmen, shepherds, and their families in order to identify cases of echinococcosis morbidity;
- strengthen sanitary inspection at the sites for content of animals and disposal of bodies of animals that have died of echinococcosis;
- keep records of dogs on farms, organize their registration, and keep track of their deworming;
- for rayon executive committees and the agricultural councils to activate state sanitary control of the agricultural and veterinary service for deworming working and guard dogs exclusively by the rules and for disposal of stray and homeless. ©COPYRIGHT: Izdatelstvo "Ala-Too", 1990

**Use of Hyperimmune Antipyocyanous Plasma in Treatment of Infectious Complications**

917C0207 Moscow *KHIRURGIYA in Russian No 11, Nov 90 pp 121-123*

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[Abstract] A study of the clinical and immunological effectiveness of hyperimmune antipyocyanous donor plasma during treatment of 27 patients with infectious complications of pyocyanous etiology after extensive heart and lung surgery involved intravenous injection of 250 ml of plasma accompanied by three to six transfusions in one to two days. Mediastinitis patients received plasma locally in addition to i.v. injections. Success of the therapy depended greatly upon the time of beginning treatment. The earlier its use after bacteriologically confirmed complications, the more successful was its use. All five cases ending in death involved a delay in beginning treatment. Early use of the treatment reduced mortality. A good clinical effect resulted from use of the treatment in complex therapy due to amplification of non-specific factors of protection of the organism and the increase of the level of specific antipyocyanous antibodies. References 11: 8 Russian, 3 Western.

**Electrostimulation of Dorsal Muscles in Walking as Method of Treating Scoliosis**

917C0388A Moscow *ORTOPEDIYA, TRAVMATOLOGIYA I PROTEZIROVANIYE in Russian No 11, Nov 90 (manuscript received 24 Apr 90) pp 1-5*

[Article by A. I. Kazmin, V. Ye. Belenkiy and A. M. Cherkashov; Central Institute of Traumatology and Orthopedics (Director—professor Yu. G. Shaposhnikov), Moscow]

UDC 616.711-007.55-08:616.741-085.844

[Abstract] Electrostimulation of the sacrospinalis and the obliquus abdominis externus muscles of the abdomen during walking, carried out at different times after closed chemonucleolysis, on 15 patients with dis-plastic lumbar scoliosis II or III degree involved the use

of a walking corrector developed at the Central Scientific Research Institute of Prostheses and Prostheses Construction. The procedure showed clearly that this form of stimulation can be useful in rehabilitation therapy after closed chemonucleolysis therapy. It also was effective in treating patients with lumbar scoliosis at different stages of the disease in a complex of conservative therapy. The persistent therapeutic effect of the procedure permitted the delay of a second course of electrostimulation for six months in contrast to the case for other methods of therapy. This made it possible to increase conservative treatment and to reduce the number of patients who need surgical intervention. References 13: 5 Russian, 8 Western.

**Use of Demineralized Bone Sawdust in Treatment of Spinal Column Injuries**

917C0388B Moscow *ORTOPEDIYA, TRAVMATOLOGIYA I PROTEZIROVANIYE in Russian No 11, Nov 90 (manuscript received 12 Jun 89) pp 22-23*

[Article by A. I. Shvets and G. I. Fadeyev; Department of Traumatology, Orthopedy and Field Surgery (head-professor—G. I. Fadeyev), Lugansk Medical Institute]

UDC 616.711-001-089:615.466]-089.168-07

[Abstract] A study of the possibility of producing inter-body bone adhesions after injection of a bone matrix into the intervertebral disk involved two series of experiments on dogs. The first experiment included exposure of the spinal column and fracture, with the aid of an osteoma, of the L<sub>1</sub> or L<sub>IV</sub> body, penetrating both disks. An osteoinductive mixture was placed in one disk and the other disk was the control. In the second series of experiments a fracture of the body, penetrating into adjacent disks, was produced. The inductive mixture was injected into one disk and the mixture and lecosime was injected into the other. Five dogs underwent each series of experiments. All preparations produced complete consolidation of fragments of the broken vertebrae with formation of a trabecular structure. Two dogs developed kiphosis at the spot of the injury and one developed aseptic necrosis of the vertebra with kephotic deformation. Osseous adhesions occurred at the level of disks into which the osteoinductive mixture was injected. Osseous adhesion occurred in one dog (20 percent of the cases) after injection of lecosime. Osteogenesis did not occur after experiments involving the control group. Clinical injection of the osteoinductive mixtures into 12 patients with remote and fresh injuries of the spine produced formation of a perivertebral osseous connection in 63.6 percent of the patients within 8-12 months. References 6: 3 Russian, 3 Western.

**Acquired Immune-Deficiency Syndrome**

917C0381A Moscow ZHURNAL MIKROBIOLOGII,  
EPIDEMIOLOGII I IMMUNOBIOLOGII in Russian  
No 8, Aug 90 (manuscript received 27 Nov 89) pp 20-25

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UDC 616.98:578.828.6]:[614:374

[Text] As long as neither reliable treatment methods nor effective vaccines against the acquired immune deficiency syndrome (AIDS) exist, educating the public on ways to prevent infection by human immune deficiency virus (HIV) is vitally important. At present, the World Health Organization assesses this as the principal method of prevention. Since 1985 the USSR has also been conducting a public health educational campaign on AIDS prevention with the participation of the mass media.

The objectives of the work have included assessing how well young people from 14 to 18 years of age are informed about AIDS, revealing attitudes toward different sources of information about AIDS, and studying the influence of lectures on the level of knowledge and the shaping of opinions on this problem. We conducted a detailed evaluation of the public's AIDS awareness earlier [2]. In this study we were interested in the level of AIDS knowledge of precisely this age group, because in the immediate future it will be the most sexually active part of the population, and depending on sexual behavior, it will be exposed to a greater or lesser danger of HIV infection by the sexual pathway.

**Materials and Methods**

The research was conducted by anonymous survey in February-April 1989. The questionnaire contained 10 questions with multiple-choice answers. The respondents were asked to select the answers that were correct in their opinion, and to indicate their age, sex and the type of educational institution they were attending.

A total of 775 questionnaires were distributed. Two groups of students from 14 to 18 years old (students of Moscow's secondary schools, medical schools and vocational-technical schools) were selected at random. Special public health education concerning AIDS prevention had not been conducted formerly in either of these groups.

In the experimental group (362 persons), the questionnaires were filled out immediately after a lecture on preventing HIV infection. In the control group (413 persons), this was done without preliminary public health education. The responses of this group were used to judge the background awareness of young people.

The effectiveness of the lectures was assessed by paired comparison of the responses of the experimental and

control groups for each response choice, while the level of knowledge was assessed from the percentage of persons believing this response to be correct.

Statistical treatment of the data entailed comparing two sampling fractions of the choices (as percentages) in the case of an alternative distribution, and Spearman's rank correlation [1,3,4]. The analysis was conducted in relation to sex, age and type of educational institution.

**Results and Discussion**

Seven hundred seventy-four completed questionnaires were received (one questionnaire was not returned). Seven hundred fifty questionnaires were analyzed (24 questionnaires were found to be unfit for analysis because the respondents were less than 14 or more than 18 years of age); of these, 359 were questionnaires from the experimental group (77 young men and 282 young women) and 391 questionnaires were from the control group (163 young men and 228 young women).

Generalization of the responses of the control group produced the following pattern:

The overwhelming majority of the respondents (94.3 percent) answering question 1 ("What is AIDS?") gave the correct response ("An infectious disease caused by a virus"), and only 1.54 percent of the respondents believed AIDS to be a noninfectious disease. This question was not answered by 4.09 percent of the respondents. Differences in the responses depending on age and sex were not found.

Analysis of responses to question 2 ("By what pathways is AIDS transmitted?") showed that the majority had the right idea about the pathways by which HIV infection is transmitted. Practically all (98.72 percent) knew about the sexual transmission pathway, 95.91 percent knew about transmission of the infection by blood, and 84.4 percent knew about transmission from mother to child during pregnancy, childbirth and nursing. To our amazement the respondents demonstrated a level of knowledge that was comparable to and even higher than that of upperclassmen in New York in late 1988 [5]. However, some of them believed that HIV is also transmitted by other pathways: by saliva when kissing (12.02 percent), by mosquito bites (8.95 percent), by perspiration (2.05 percent), by handshake (0.77 percent), and in baths and swimming pools (0.77 percent). The responses do not depend on the age and sex of the respondents.

Generalization of the responses to question 3 ("Who is in the greatest danger of being infected by AIDS?") revealed that the overwhelming majority knew about risk groups. In the opinion of the participants of the survey, those in greatest danger of AIDS infection include prostitutes (97.95 percent), people who change their sex partners often (95.40 percent), persons who have sexual contact with ignorant people (95.14 percent) and with foreigners (92.07 percent), bisexuals (91.05

percent), drug addicts (89 percent), homosexuals (80.56 percent), and people who receive blood transfusions frequently (73.15 percent).

The students placed bisexuals in risk groups more often than homosexuals. They possibly believed that bisexuals expose themselves to a double danger of AIDS infection because they could catch the virus both in the homosexual environment and from women. Moreover some of the respondents maintained the opinion that it was also dangerous to be a donor (65.22 percent), to frequent hair salons (7.93 percent) and baths and swimming pools (1.3 percent), and to dine in public places (1.02 percent).

Over half of the respondents felt donating blood to be dangerous. It must be said that although we phrased the question in a way to preclude an elaborate response, the respondents commented on it extensively anyway. An analysis of these additional responses provided an impression of the motives of the respondents for placing blood donors in the risk group ("only in the USSR," "in our country"). When interviewed, the students explained their placement of donors in the risk group by their uncertainty that blood is always taken with sterile instruments and by their mistrust of the lecturer's statements that blood is taken only with disposable instruments. It may be that placement of donors in the risk group was also influenced by the fact that donors in our country are subject to mandatory AIDS testing, and they are often referred to in printed material in the same breath as risk groups.

Age and sex analysis of the responses to this question shows that presence of a dependable correlation ( $p > 0.1$ ) was not established between the age of the respondents and the frequency of "positive" responses, while the influence of the sex of the respondents was revealed in relation to only one response choice: It was primarily young women who felt that people who frequent hair salons face the danger of AIDS infection ( $p < 0.05$ ). Such a dependence is obviously explained by the fact that young women associated visits to hair salons with manicures and pedicures, while young men associated them most often only with haircuts. Comments made by young women provide evidence of this: "manicures, poorly processed instruments (blood still on them)," "manicures, pedicures."

The responses to question 4 ("What must be done to avoid AIDS infection?") showed that the majority of the respondents were aware of measures by which to prevent HIV infection. The need for using a condom in random sex encounters was indicated by 90.26 percent of the respondents, the need for intimacy only with a constant sex partner was stated by 89.49 percent, avoiding injection of narcotic drugs was stated by 86.92 percent (some added: "With an unsterilized syringe," "Syringes must be sterilized well," "Disposable syringes should be used," "Don't make injections with the same syringe and needle"), and not using the services of private individuals for medical procedures (injections, tattoos, abortions, ear piercing etc.) was stated by 73.08 percent.

Part of them felt that AIDS infection can also be prevented by not donating blood (35.9 percent), by avoiding mosquito bites (8.46 percent), by not kissing (2.82 percent), by not going to baths, swimming pools and hair salons (2.05 percent), by not using public toilets (1.8 percent), and by not drinking carbonated water from automatic dispensers (1.54 percent).

It is interesting that 65.22 percent of the respondents placed donors in risk groups, while only 35.9 percent gave "not donating blood" as a way to prevent AIDS infection. The older the respondents, the more frequently they stated the need for using condoms to prevent HIV infection and the less danger they saw in donating blood.

As for question 5 ("Can just a single sexual contact cause AIDS infection?"), 94.1 percent of the respondents answered correctly, 2.57 percent gave the wrong answer, and 3.33 percent answered "I don't know." The frequency of correct responses increased with age. Young men gave the wrong answer more often ( $p < 0.05$ ).

In response to question 6 ("From what sources have you received AIDS information?"), most named several sources (an average of 3.8 sources per person). The sources of information fell into the following decreasing order: television (96.4 percent), newspapers and magazines (90.75 percent), discussions with acquaintances (52.96 percent), pamphlets and books (46.53 percent), lectures (32.65 percent), other sources (25.96 percent), talks with doctors (17.48 percent) and posted newspapers (14.91 percent).

Sex differences were not detected in this distribution; however, while both young women and young men ranked discussions with acquaintances third as a source of information, young women named this source significantly more often (young men—45.34 percent, young women—58.33 percent). A dependence of the responses on age was also established: The older the respondents, the more information they received from books, pamphlets and posters and the less from discussions with acquaintances.

Over half of the respondents named discussions with acquaintances among the information sources, and this source was noted significantly more often than all of the others, except for television and the press. This is an indication that this problem is being discussed actively among young people, but it also suggests that other information sources are either not very accessible or are uninteresting. This conclusion is also confirmed by responses to question 7 ("From what sources do you prefer to obtain information about AIDS?"). "Discussions with acquaintances" shifts as an information source from third to sixth place. The distribution of information sources from which respondents would like to receive information about AIDS is: television (91.24 percent), newspapers and magazines (85.31 percent), talks with doctors (68.04 percent), lectures (66.24 percent), pamphlets, books and posters (49.74 percent),

discussions with acquaintances (25.26 percent), other sources (17.27 percent) and posted newspapers (13.66 percent).

The respondents prefer to obtain information from several sources. There is an average of 4.2 sources per respondent. Among them, respondents named radio and video tapes, and they expressed a desire to obtain information from carriers of the virus themselves ("if they are not opposed to doing so"). Sex differences were not detected in the responses. The following pattern was revealed in an age analysis of the responses: Greater preference is shown with age to books, pamphlets and posters, and correspondingly less is shown to lectures ( $p < 0.05$ ). Television, newspapers and magazines take the lead in all age groups, and posted newspapers are at the bottom of the list just as consistently. Obviously students are not satisfied by the informativeness and the format of the public health bulletins and posted newspapers they have encountered.

Comparison of the responses to questions 6 and 7 provides the grounds for concluding that the need of young people for information on the AIDS problem is not being fully satisfied. This conclusion is also confirmed by responses to subsequent questions on the questionnaire.

Most respondents gave a positive response to question 8 ("Would you wish to have printed information on AIDS in your own home?"). The sex distribution was a factor significantly influencing the responses: 72.49 percent young men and 85.16 percent young women ( $p < 0.001$ ).

Approximately the same relationship was revealed in responses to question 10 ("Do the mass media report enough about AIDS?"). Only 36.65 percent of young

men and 21.33 percent of young women felt this information to be sufficient ( $p < 0.001$ ), while the rest felt it to be insufficient.

Young men and women also related differently to their knowledge of the AIDS problem. The assessment given to their knowledge was "very high" for 5.52 percent of the young men and only 1.32 percent of the young women, "high" for 13.50 and 9.25 percent, "average" for 72.40 and 74.12 percent, and "low" for 8.59 and 14.91 percent. Consequently young men gave a higher assessment to their AIDS knowledge than young women. There were more young men among those who felt AIDS information communicated by the mass media to be sufficient, while significantly fewer young men would want to have printed AIDS information at home. But the answers to questions 1-8 show that the level of knowledge of young women is approximately the same as that of young men, while in regard to question 5 it was the young men who had the wrong impression most often. Thus the difference in self-assessment of knowledge does not reflect the true situation. But obviously a higher self-assessment given of their knowledge by young men is associated with the fact that they display less interest in printed information, and that they are less demanding of the mass media than are young women.

The questionnaire survey carried out in the experimental group, which was subjected to preliminary public health education taking the form of lectures, showed that significant differences exist in comparison with responses of the control group only in relation to some of the responses. But on the whole, the opinion of this group appears more realistic.

The results of comparing the responses of the experimental and control groups are given in the table (questions 1-5, 10).

Questionnaire Results (Percent "Positive" Responses)

Question Number	Question and Response Choices	Group		p
		Control (n = 391)	Experimental (n = 395)	
1	<i>What is AIDS?</i>			
	An infectious disease caused by a virus	94.37	99.44	<0.001
	A noninfectious disease	1.54	0.28	<0.1
	No response	4.09	0.28	[0].001
2	<i>By what pathways is AIDS transmitted?</i>			
	By handshake	0.77	0.28	>0.1
	By mosquito bites	8.95	2.79	<0.001
	From mother to child during pregnancy, childbirth and nursing	84.40	95.53	<0.001
	Through the air during conversation	0.77	0.28	>0.1
	Sexually	98.72	98.88	>0.1
	By blood	95.91	98.32	<0.05
	By baths, swimming pools	0.77	0.84	>0.1
	By saliva when kissing	12.02	6.15	<0.05

Questionnaire Results (Percent "Positive" Responses) (Continued)				
Question Number	Question and Response Choices	Group		p
		Control (n = 391)	Experimental (n = 395)	
	By perspiration	2.05	1.96	>0.1
3	<i>Who is in the greatest danger of being infected by AIDS?</i>			
	Men living a sexual life with other men	80.56	96.65	<0.001
	Men living a sexual life with both men and women	91.05	96.65	<0.001
	People dining frequently in public places	1.02	0.84	>0.1
	Prostitutes	97.95	96.65	>0.1
	Blood donors	65.22	60.34	>0.1
	People entering into sexual contact with foreigners	92.07	94.69	>0.1
	People visiting baths and swimming pools frequently	1.30	1.39	>0.1
	Drug addicts	89.00	93.86	<0.5
	People receiving blood transfusions frequently	73.15	77.37	>0.1
	People changing sex partners frequently	95.40	96.65	>0.1
	People visiting hair salons frequently	7.93	6.70	>0.1
	People entering into sexual contact with ignorant people	95.14	96.09	>0.1
4	<i>What must be done to avoid AIDS infection?</i>			
	Don't drink carbonated water from automatic dispensers	1.54	1.40	>0.1
	Live a sexual life only with a constant sex partner	89.49	89.39	>0.1
	Don't kiss	2.82	0.56	<0.05
	Use a condom in random sexual encounters	90.26	93.30	>0.1
	Don't inject narcotic drugs	86.92	86.03	>0.1
	Don't go to baths, swimming pools, hair salons	2.05	2.24	>0.1
	Don't use the services of private persons for medical procedures (injections, tattoos, abortions, ear piercing etc.)	73.08	82.96	<0.01
	Don't use public toilets	1.80	1.68	>0.1
	Don't donate blood	35.90	30.73	>0.1
	Avoid mosquito bites	8.46	2.514	<0.001
5	<i>Can just a single sexual contact cause AIDS infection?</i>			
	Yes	94.10	93.32	>0.1
	No	2.57	2.24	>0.1
	Don't know	3.33	4.44	>0.1
10	<i>Do the mass media report enough about AIDS?</i>			
	Yes	27.87	16.47	<0.001
	No	72.14	83.52	<0.001

There were significantly more correct responses to the question "What is AIDS?" in the group attending the lecture.

Judging from the survey, this group had a truer idea about the pathways of HIV infection transmission. The respondents referred much more often to transmission pathways like "by blood" and "from mother to child," and wrong answers were rarer. Three times fewer respondents believed that AIDS is transmitted by mosquito bites, and twice fewer saw such danger in kissing.

The bulk of the respondents placed homosexuals, bisexuals and drug addicts in risk groups. While in the control group sex differences were not detected in relation to the answer "people changing sex partners frequently," such a dependence was established in the experimental group: Young women placed these people in risk groups more often than young men. If we compare the responses of young men in the experimental and control groups, we find that they do not differ significantly, while young women who attended the lecture placed these people in risk groups more often than did young women of the control group (98.22 and 94.74 percent respectively,  $p < 0.05$ ).

Respondents of the experimental group (to question 4) placed a higher value ( $p < 0.01$ ) on "don't use the services of private persons for medical procedures (injections, tattoos, abortions, ear piercing etc.);" as a way of preventing HIV infection. They also saw less danger in kissing ( $p < 0.05$ ) and in mosquito bites ( $p < 0.001$ ).

While the position of young men and women in the control group in relation to the mass media from which the respondents preferred to obtain AIDS information coincided, opinions diverged in the experimental group. Young men adhered to the same opinion as their peers in the control group, while young women moved the source "newspapers and magazines" from second to fourth place, and lectures from fourth to second.

Respondents of the experimental group were more critical of reports on AIDS in the mass media. These reports were insufficient for a significantly larger number of them in comparison to the control group; moreover a dependence upon the sex of the respondents was also observed in this group. Only 21.53 percent of the young men and 15.61 percent of the young women felt that the mass media report on AIDS sufficiently, while for the rest, this information was insufficient.

Analysis of the responses in relation to types of educational institutions showed that the opinion of students of secondary schools, medical schools and vocational-technical schools do not differ significantly.

Thus significant differences were discovered between the levels of knowledge of the group that attended the lecture and the control group. However, mistaken opinions were observed in relation to a number of questions even after attendance of the lecture. Obviously constant explanatory work on prevention of HIV infection making more

effective use of propaganda resources is required. Special attention should be devoted in public education to explaining that HIV is not transmitted by blood donation, by mosquito bites, by kissing and by perspiration.

Although the background level of awareness of the 14 to 18 year age group is rather high, we feel that a gap exists between the level of knowledge and practical implementation of this knowledge. We will try to analyze this problem in subsequent papers.

### Conclusions

1. Young people 14 to 18 years old possess a relatively high level of knowledge on the pathways by which HIV infection spreads.
2. Television and the press have the greatest influence on the level of AIDS knowledge possessed by young people.
3. Lectures are a resource making it possible to correct the AIDS knowledge of young people obtained from nonmedical sources.

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### Meadow-Field Type of Tularemia Foci in Eastern Siberia

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[Text] Three types of tularemia foci have been known up until now in Eastern Siberia: floodplain-marsh, steppe and tundra [1-3]. At the same time, the nature of the natural conditions in Eastern Siberia, and particularly in southern agricultural regions, provided the grounds for hypothesizing the presence of meadow-field foci here as well. This hypothesis was confirmed in 1985, when a diffuse tularemia epizooty, which arose on the background of a high abundance of Eastern European field mice, was recorded in Uzhurskiy Rayon, Krasnoyarsk Kray [4]. A tularemia focus revealed itself for the first time in 1988 in Zalarinskiy Rayon, Irkutsk Oblast<sup>1</sup> as one of significant epizootic activity never noted previously in the Baikal region. The high abundance of mouse-like rodents noted as early as in the beginning of winter 1987-1988 and its subsequent extensive participation in the epizooty predetermined the need for emergency epidemic control measures, and particularly, vac-

cination of the rayon's population, which in turn necessitated study and determination of the typology of this focus.

#### Materials and Methods

The principal research was conducted in two areas approximately 18 km apart and separated by the watershed of two small rivers—the Unga and the Zalarinka, in the vicinities of the villages of Bolshaya Zaimka and Sorty. Commonly accepted zoological and parasitological methods were used to make the surveys. Field material was delivered to a laboratory in Dewar flasks and analyzed biologically using biological tests on white mice. In the period from the end of April to the end of September 803 rodent and insectivore specimens, 351 specimens of blood-sucking arthropods and 28 water samples were tested bacteriologically, as a result of which 18 strains of *Francisella tularensis* were isolated (see table).

Results of Bacteriological Testing of Field Material From Zalarinskiy Rayon for Tularemia in 1988

Date Material Collected	Type of Material	Number of Specimens, Samples Tested	Number of Biological Tests	Number of Tularemia Agent Cultures Isolated
19 Apr - 29 Apr	Corpses of <i>Microtus gregalis</i>	54	7	7
	<i>Microtus gregalis</i>	103	7	7
	<i>Microtus oeconomus</i>	8	1	-
	Red-backed voles	2	1	-
	Shrews	6	3	-
21 Jun - 4 Jul	Corpses of <i>Microtus gregalis</i>	12	12	2
	<i>Microtus gregalis</i>	235	19	1
	East Asian mice	9	3	-
	Ticks	191	8	-
	Fleas from <i>Microtus gregalis</i>	120	3	-
	Lice	40	1	-
	Water from flooded streams of the Unga river basin	4 samples	4	1
	Water from the Unga River	8 samples	8	2
12 Aug - 22 Aug	Water from the Zalarinka River	2 samples	2	-
	<i>Microtus gregalis</i>	111	13	2
	East Asian mice	24	5	-
	Red-backed voles	4	1	-
	<i>Lepus timidus</i>	1	1	-
12 Sep - 22 Sep	Shrews	36	6	-
	<i>Microtus gregalis</i>	134	11	-
	East Asian mice	16	4	-
	Red-backed voles	11	3	-

	Reddish gray voles	2	1	-
	Field mice	4	2	-
	Shrews	31	5	-
	Water from flooded streams of the Unga river basin	7 samples	7	-
	Water from the Unga River	3 samples	3	-
	Water from the Zalarinka River	4 samples	4	-

## Results and Discussion

The focus described here is at the boundary between the Balagan forest-steppe and sub-taiga pre-Sayan forest. In natural respects its territory is a typical agricultural landscape with a hilly topography. Plowed fields are interspersed primarily with small timber stands and forest outliers made up of birch, aspen and pine, confined as a rule to the summits of hills and their steep slopes. Low areas are occupied by sedge-hummock marshes of small area. The floodplains of the Zalarinka and Unga rivers and of the streams flowing into them are extremely marshy in places. Steppe-invaded areas are occupied by territory unsuited for plowing. The farmland is used primarily to grow cereal crops. Water is supplied to population centers primarily from artesian wells.

According to census data the small-mammal population is represented by red-backed [krasnaya] and reddish gray [krasno-seraya] voles, by the vole *Microtus gregalis* [uzkocherepnaya], *Microtus oeconomus*, field and East Asian mice, harvest mice and shrews. Steppe regions, boundary strips and roadsides are inhabited by long-tailed susliks, while squirrels and hares (*Lepus timidus* and *L. europaeus*) inhabit forested areas. Besides the species listed above, muskrats are also encountered. But their population is small because of unfavorable natural conditions. According to reports from local inhabitants water rats live in the area. Ixodid ticks include two species: *Ixodes persulcatus* P. Sh. and *Dermacentor nuttalli* Ol. The former inhabits forest outliers and forested river floodplains, while the latter inhabits steppe regions, roadsides and perennial grass plantations. Eleven species of fleas, three species of lice and 12 species of Hama-did ticks were recorded on trapped animals.

During the survey (which began in December 1987) 924 specimens of mouse-like rodents were trapped within the areas indicated above, including 694 specimens (75 percent) of *Microtus gregalis*. In early June 1988 the abundance of the latter was unusually high for this time, averaging 40 percent in 100 trap-nights in different habitats. By the end of spring 1988, *Microtus gregalis* inhabited practically all stations noted in the territory of the focus. The abundance of other rodents was noticeably lower, being 18 per 100 trap-nights for East Asian mice, up to seven for red-backed voles and up to three for field mice in June 1988. Single specimens of the reddish gray vole were caught. Nine percent of the total

quantity of trapped animals were shrews. Practically no *Microtus oeconomus* were caught in 1988. It is important to note that water rats were detected in the survey territory in neither 1987 nor 1988.

The first manifestations of an already proceeding, intensive tularemia epizooty were noticed in the vicinity of the village of Bolshaya Zaimka in the second half of April 1988, after disappearance of snow cover, when corpses of *Microtus gregalis* were discovered in bunches of straw and in natural localities. Seven cultures of tularemia agent were isolated from the corpses of these animals found from 19 to 20 April. Another five cultures were isolated from live *Microtus gregalis* caught during this time. Water of the Unga River and of some bodies of water communicating with its basin was also found to be infected.

From late May to early June 1988 the epizooty went into a decline, and it ceased by August, which is confirmed by the results of bacteriological analysis of field material. It obviously began in December 1987 or January 1988. Intensive reproduction of *Microtus gregalis* was noted during this time. Animals were concentrated in fields, in piles of uprooted timber, bunches of straw and haystacks. Judging from everything, the maximum of epizootic activity occurred around the second half of the winter and the beginning of spring.

Thus the epizooty affected only *Microtus gregalis*, and it arose in winter. If rodents of other species were drawn into the epizootic process, it was sporadically, as a result of which we were unable to reveal infected specimens among them.

The pronounced seasonality of the focus's activity, which coincides with the winter period, and participation of *Microtus gregalis* in the epizootic process as the principal carrier of infection—a typical representative of the agricultural landscape of the southern regions of Eastern Siberia—provide the grounds for classifying this focus as being of the meadow-field type. In contrast to European meadow-field foci, in which infection is maintained primarily in populations of the common vole, this focus functions due to *Microtus gregalis*. This circumstance allows us to treat it as a Siberian variant of meadow-field foci.

The focus was also highly active from an epidemiological standpoint. Two tularemia patients were registered by

the time vaccinations of the population of Zalarinskiy Rayon. In one case a female inhabitant of the village of Troitska working as a calfmaid at a farm of Vlast Sovetov Kolkhoz fell ill on 19 May. The patient was in constant contact with hay and straw from an area in which many corpses of *Microtus gregalis* killed by tularemia were collected during the survey in June, which was confirmed by isolation of cultures of the agent of this infection from them. The illness progressed with development of a bubo in the right axillary region, appearing in the third day of illness. A skin test taken on the 13th day after the beginning of illness was sharply positive, the agglutination reaction with tularemia diagnosticum was 1:400, and the passive hemagglutination reaction was 1:1,600. In another case a department director of Khor-Tagninskiy Sovkhoz fell ill on 11 June. Painfulness and swelling appeared in the region of the left submaxillary lymph nodes on the third day of illness. The diagnosis was confirmed by a positive skin test and a titer of 1:200 in the agglutination reaction using tularemia diagnosticum.

Illness had a severe onset in both cases, with chills and high temperature, attaining 39-42°C. The subsequent clinical pattern was typical of the bubonic and anginous-bubonic forms of tularemia. Infection probably occurred in the first case by hay or straw infected by the agent, and by water in the second case.

The classification of the focus described above, and of the focus revealed in 1985 in Krasnoyarsk Kray, raises no doubts, and it provides the grounds for stating that presence of meadow-field tularemia foci in Eastern Siberia is an established fact. The genesis of these foci is obviously similar to that of European meadow-field foci, and it is associated with development of forest areas for farming. A typical feature of the foci is that diffuse epizootics develop in them apparently only on the background of a high abundance of *M. gregalis* and *Microtus subarvalis* (presence of the latter was established in Irkutsk Oblast). At the same time, massive reproduction of these animals is observed to be separated by sizable time intervals, sometimes of several years. This circumstance complicates epidemiological surveillance of the activity of meadow-field foci to a certain degree, and predetermines the need for organizing the monitoring of the dynamics of *M. gregalis* and *M. subarvalis* populations in agricultural landscapes of the south of Eastern Siberia. In this case, in years of sharp growth of their abundance there must be greater control over the sanitary condition of public water supplies and over the recycling of straw that always remains in the fields after harvesting, and rat eradication measures must be activated at animal farms and complexes, and when necessary, at other facilities as well.

#### Conclusions

1. Presence of meadow-field tularemia foci was established in farming regions of the south of Eastern Siberia. *M. gregalis* and *M. subarvalis* are the principal carriers in them.

2. The greatest epizootic activity occurs in the foci in winter or early spring on the background of a high abundance of the principal carriers of infection and their concentration on plowed fields, in straw piles, and in haystacks and hayricks.

3. People may be infected in meadow-field foci of Eastern Siberia by contact with straw and hay, as well as by water from open-air water sources infected by sick rodents.

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#### Footnote

1. An epidemiological post for long-term epizootological observations was organized here in 1987 by the epidemiological station. ©COPYRIGHT: Izdatelstvo "Meditsina", 1990

#### Clinical and Epidemiological Features of the Course and Diagnosis of North Asian Tick-Borne Rickettsiosis

917C0381C Moscow ZHURNAL MIKROBIOLOGII, EPIDEMIOLOGII I IMMUNOBIOLOGII in Russian No 8, Aug 90 (manuscript received 20 Nov 89) pp 47-49

[Article by G. S. Tomilka, G. S. Kovalskiy and I. S. Starostina, Khabarovsk Medical Institute]

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[Text] Intensive industrial development of sparsely inhabited territories of Siberia and the Far East and the advent of new population centers are doubtlessly widening man's contact with nature, and can promote revelation of previously unknown foci of tick-borne rickettsiosis, and naturally, growth of morbidity. This is the reason for the greater interest in the problems of timely diagnosis, comprehensive treatment and prevention of tick-borne rickettsiosis.

The objective of this work was to study the clinical and epidemiological features of the course of tick-borne rickettsiosis today, and to develop additional methods of its diagnosis.

### Materials and Methods

Eighty-eight tick-borne rickettsiosis patients (27 women and 61 men) were under observation. One patient was 15 to 20 years old, 59 were 21 to 50 years old, and 28 patients were over 50. Thirty patients had the mild form of the disease, 56 had the moderately severe form, and two had the severe form. All patients were undergoing antibiotic therapy with tetracycline or levomycetin. No deaths were observed among the patients.

Besides the widely employed complement fixation reaction, we used the leukocytolysis reaction, as modified by us (efficiency proposal No 769 dated 7 January 1984, Khabarovsk Medical Institute), and the agar precipitation reaction (efficiency proposal No 761 dated 15 November 1983), for laboratory diagnosis of tick-borne rickettsiosis.

### Agar Precipitation Reaction Procedure

Immune complexes were obtained by diluting 0.6 ml blood serum in borate buffer solution (3.41 gm boric acid and 4.275 gm borax in 1 ml distilled water, pH 8.4) at a 1:3 ratio. An equal quantity of 7 percent polyethylene glycol solution (molecular weight 6,000), prepared with borate buffer solution, was added to the resulting diluted serum. The mixture was incubated for one hour at room temperature. Then it was centrifuged for 10 minutes at 3,000 rpm, the supernatant was drained off, and the precipitate remaining in the test tube was washed twice with 3.5 percent polyethylene glycol solution and centrifuged for 10 minutes at a time at 3,000 rpm. The resulting precipitate was dissolved in 0.05-0.1 ml 0.01 N sodium hydroxide solution.

In order to detect specific antibodies in precipitated immune complexes, Sibirikus, a dry rickettsiosis antigen produced for the complement fixation reaction, was dissolved in physiological solution of sodium chloride to obtain a titer four times exceeding that indicated on the ampule. A 4 ml batch of antigen was prepared by heating in a water bath to 56°C. Then 4 ml Difko agar produced by Ferak (GDR), melted and cooled to 56°C, was added to this quantity of antigen. The resulting mixture of agar and Sibirikus rickettsiosis antigen was poured onto a clean 9x12 cm piece of glass positioned horizontally. Craters with a 4 mm diameter were punched out of the solidified agar 20-30 minutes later. The solution of immune complexes was transferred to these craters with a capillary or a Pasteur pipette, filling them even with the agar surface. After the craters will filled, the glass bearing the agar was placed in a humidified chamber at room temperature. The reaction was checked after 36-48 hours. A distinctly outlined zone of precipitation indicated presence of antibodies in the immune complexes of blood serum. To permit long-term storage of the reaction results, the plates bearing the agar were washed in physiological solution of sodium chloride and then dried for one day and stained with a 0.1 percent solution of 10B amido black in 10 percent acetic acid. Presence of

precipitation zones in agar containing Sibirikus rickettsiosis antigen, coupled with an increase in their diameter in a second batch of serum, was the basis for diagnosing tick-borne rickettsiosis.

### Leukocytolysis Reaction Procedure

Heparinized blood (2 units of heparin per 0.1 ml blood) was added to two test tubes at a quantity of 0.1 ml each. Then 0.1 ml physiological solution of sodium chloride was added to one of the test tubes (the control), and 0.1 ml of Sibirikus rickettsiosis antigen, produced for the complement fixation reaction and initially diluted with physiological solution of sodium chloride in the amount indicated on the ampule, was added to the second (experiment). After two hours of incubation at 37°C (with periodic shaking), 0.02 ml of blood were taken from each test tube and transferred to two test tubes containing 0.4 ml 3 percent acetic acid solution, in which destruction of erythrocytes occurred. The number of blood leukocytes was counted by the usual procedure in Goryayev counting chambers. The reaction was evaluated by calculating the difference between the leukocyte counts in the control sample  $M_c$  and in the sample to which antigen had been added ( $M_e$ ), expressing it as a percentage of the control sample, and it was assessed as positive when the leukocytolysis factor (LF) was 10 percent or more:

$$LF = [(M_c - M_e)/M_c] \times 100\%.$$

### Results and Discussion

A diagnosis of tick-borne rickettsiosis was made on the basis of data from a clinical examination, together with mandatory consideration of the epidemiological medical history, and it was reinforced with laboratory data.

An acute beginning of illness was typical, having been established in 69 out of 88 examined patients with tick-borne rickettsiosis. Intoxication was accompanied by headaches in all patients. The fever lasted from three to 14 days. Because there were basically no patients with a severe course among those hospitalized, the longest duration of fever was recorded among patients with the moderately severe form of tick-borne rickettsiosis (an average of 8 to 9 days). A drop in temperature in the course of 1 to 2 days in response to tetracycline was typical. The fact that tetracycline is so highly effective against tick-borne rickettsiosis can be used in diagnosing this illness.

The primary response at the tick attachment location was a pathognomonic symptom of tick-borne rickettsiosis. It occurred in the majority of patients (72.9 percent).

Enlargement of peripheral regional lymph nodes was noted in 25 percent of the patients within the zone of entry of the infection.

The cardinal symptom of disease—a rash—was observed on all patients. Clinical observations of recent years indicate that the number of patients among whom a rash appears a longer time after the beginning of illness has increased in comparison with the 1960s (from 24.6 to 53.6 percent) [1]. Moreover the rash was often relatively mild, and less pronounced (47.8 percent). Among other features of the course of tick-borne rickettsiosis today, mention should be made of the tendency for the rash to concentrate on the torso and the upper limbs (39.6 percent). The rash was of the roseola-papular type. Individual elements attained a diameter of 10 mm, and as a rule they did not merge with each other. Changes on the part of other organs were insignificant and nonspecific in the presence of tick-borne rickettsiosis.

The epidemiological history had important significance to early identification of tick-borne rickettsiosis. Establishment of the fact of visiting wooded and brushy areas in the spring-summer period within 14 days of falling ill was decisive. Tick-borne rickettsiosis occurred more frequently among men (69.3 percent) and persons 20 to 50 years old (67 percent). In recent years, however, the number of tick-borne rickettsiosis patients in the older age group increased (31.8 percent). This is the result of considerably higher interest in maintaining summer cabins. The number of cases of tick-borne rickettsiosis among urbanites increased noticeably in comparison with the rural population.

The characteristic clinical symptoms of tick-borne rickettsiosis and its epidemiological features made it possible to establish the diagnosis of this disease on the basis of clinical and epidemiological data in typical cases. At the same time presence of certain symptoms similar to those occurring with other infectious diseases required laboratory testing. Specific serological tests were found to be the most informative. The complement fixation reaction, so widely used in practice, was positive in 78 percent of the cases. The agar precipitation reaction, as modified by us [2], was found to be positive among 85.7 percent of patients. The reaction's high sensitivity is apparently the product of the greater concentration of antibodies in immune complexes precipitated from a relatively large quantity of blood serum. The proposed method is less laborious in comparison with the complement fixation reaction, it does not require fresh ram erythrocytes or guinea pig complement, tests can be run simultaneously on up to 35 blood sera, and the glass plates bearing the results can be stored long-term after appropriate processing.

The need for repeating serological reactions after two weeks is one of their significant shortcomings, excluding them from use as resources of early diagnosis. Therefore we used our modified leukocytolysis reaction to permit earlier diagnosis of tick-borne rickettsiosis [3]. The reaction becomes positive as early as in the first day of

illness, and its results can be obtained 2.5-3 hours after the reaction. Advantages of the reaction are that it is easy to run and its specificity is rather high (72.4 percent). The leukocytolysis reaction with *Sibirikus* rickettsiosis antigen can be used successfully for quick diagnosis of tick-borne rickettsiosis.

#### Conclusions

1. Features of the course of North Asian tick-borne rickettsiosis today include a higher proportion of urban residents and older persons in the disease structure, which, when compounded by the increase in number of patients with late appearance of the rash, makes early diagnosis of this disease more difficult.
2. Using a modified agar precipitation reaction and the leukocytolysis reaction is recommended as a means of improving laboratory verification of tick-borne rickettsiosis, in addition to the complement fixation reaction.

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#### Mortality Caused by Infectious Diseases in the USSR

917C0381D Moscow ZHURNAL MIKROBIOLOGII, EPIDEMIOLOGII I IMMUNOBIOLOGII in Russian No 8, Aug 90 (manuscript received 17 Jul 89) pp 53-57

[Article by I. L. Shakhanina, O. M. Ivliyeva and M. I. Narkevich, Central Scientific Research Institute of Epidemiology, USSR Ministry of Health, Moscow]

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[Text] The value of mortality information is determined not only by its role in arriving at a dependable assessment of public health and of an epidemiological situation, but also by its fundamental significance to implementing medical, social and economic programs.

When we study the trends and causes of mortality in the population, we usually focus on the classes and forms of pathology primarily responsible for its overall level. As a consequence, not enough attention has been devoted in recent years to analyzing mortality caused by infectious diseases, which make up only 2-3 percent of all causes in the structure of mortality (1970-1987). This paper attempts to fill this omission.

### Materials and Methods

We used official statistics on mortality in the USSR and in the union republics for 1970-1987 (Form No 5, USSR Central Statistical Administration).

The analysis was carried out on nosological forms of disease in class I, "Infectious and Parasitic Diseases," of the International Nomenclature and Classification of Diseases, Injuries and Causes of Death (MKB-9).

The materials were processed with an IBM PC XT personal computer using a Supercalc-4 program.

### Results and Discussion

The period in question was characterized by a tendency for growth of mortality due to all causes at an annual rate of +1.43 percent.

In this situation, special mention should be made of the fact that mortality due to class I diseases decreased from 26.6 in 1970 to 21.2 in 1987 (per 100,000 population)—that is, by 20.3 percent, at a mean annual rate of -0.44 percent. At the same time a year-by-year analysis indicates that mortality caused by infectious and parasitic diseases increased beginning in 1975, resulting in a 13.2 percent increase by 1983 (from 21.8 to 25.1 per 100,000 population).

Mortality due to infectious and parasitic diseases was 1.5-2 times higher among rural residents than urban residents. When the indicators for the urban and rural populations were standardized with respect to age, this ratio did not change.

The structure of mortality caused by infectious and parasitic diseases is determined, as follows from the name of this class, by two groups of illnesses, with the former (infectious diseases) prevailing at 99 percent. Five or six nosological forms dominate this group on an annual basis as a rule, occupying over 90 percent of the structure of class I. In 1970 they were tuberculosis (75 percent), septicemia (6.7 percent), viral hepatitis (3.3 percent), measles (2.5 percent) and dysentery (2.9 percent). Eighteen years later, in 1987, as a result of significant changes in the epidemiological situation of a number of infectious diseases, this structure was determined by tuberculosis (36.7 percent), acute intestinal infections (AII's) (29.7 percent), septicemia (10.9 percent), viral hepatitis (10.6 percent) and meningococcal infections (3.9 percent). The proportion of deaths caused by measles (1.6 percent) and salmonellosis (1.5 percent) continues to be noticeable. The relative weight of the other nosological forms in class I totals only 5.1 percent.

Mortality per 100,000 population due to individual infectious diseases is shown in Table 1.

**Table 1. Mortality Caused by Infectious Diseases in the USSR, per 100,000 Population**

Infectious Diseases	Year												
	1970	1972	1974	1976	1978	1980	1981	1982-	1983	1984	1985	1986	1987
Typhoid fever and paratyphoid	0.16	0.09	0.12	0.11	0.08	0.08	0.10-	0.08	0.11	0.08	0.07	0.05	0.05
Other salmonellosis	0.03	0.05	0.10	0.24	0.49	0.47	0.53-	0.49	0.52	0.41	0.37	0.33	0.32
Shigellosis	0.73	0.70	0.82	0.75	0.47	0.43	-0.31	0.24	0.28	0.29	0.24	0.20	0.18
AII	0.22	0.21	0.79	1.36	1.84	3.17	5.92	-6.28	7.43	6.97	6.73	6.45	6.29
Viral hepatitis	0.88	0.74	0.71	0.87	0.82	0.81	1.28	-1.31	1.66	1.59	1.82	1.87	2.24
Acute poliomyelitis	0.01	0.01	0.02	0.03	0.03	0.02	0.02-	0.03	0.02	0.2	0.01	0.01	0.02
Diphtheria	0.05	0.02	0.01	0.01	0.01	0.01	0.01-	0.01	0.02	0.03	0.03	0.02	0.02
Measles	0.66	0.29	0.31	0.22	0.24	0.50	0.39-	0.39	0.51	0.19	0.23	0.39	0.34
Whooping cough	0.03	0.03	0.01	0.03	0.02	0.01	0.02	0.02-	0.01	0.01	0.02	0.01	0.02
Scarlet fever	0.03	0.01	0.01	0.01	0.01	0.01	0.01	0.01-	0.01	0.01	0.01	0.01	0.01
Meningococcal infection	0.26	0.63	0.57	0.64	0.66	0.75	0.84	-0.91	0.98	1.07	0.99	0.85	0.82
Brucellosis	0.02	0.01	0.001	0.01	0.01	0.003	-0.004	0.005	0.01	0.01	0.01	0.01	0.01
Anthrax	0.004	0.004	0.001	0.007	0.003	0.002	-0.002	0.003	0.004	0.001	0.001	0.002	0.001

Rabies	0.02	0.03	0.04	0.03	0.03	0.03	0.02	-0.02	0.02	0.02	0.02	0.02	0.01
Tetanus	0.12	0.13	0.12	0.09	0.08	0.06	0.07	0.07	0.07	0.06	0.05	0.05	0.04
Septicemia	1.77	2.29	2.71	3.96	4.13	4.07	3.66	3.36	3.12	2.82	2.49	2.30	2.30
Syphilis	0.15	0.13	0.09	0.08	0.06	0.05	0.04	0.03	0.03	0.02	0.02	0.02	0.01
Tuberculosis	19.98	17.72	14.18	13.13	11.66	11.67	10.02	9.77	9.44	9.59	9.38	7.93	7.77
Other	0.60	0.72	0.47	0.52	0.56	0.65	0.76	-0.58	0.86	0.84	0.77	0.68	0.70
Total for class I	26.63	24.43	21.67	22.66	21.74	23.30	24.38	-29.98	25.08	24.04	23.29	21.21	21.16

In 1987 the highest mortality was noted in relation to tuberculosis—7.8. At the same time significant successes were attained during the period in question in the fight to prevent deaths due to this disease: Mortality was 2.5 times lower in 1987 than in 1970.

AII's are in second place among causes of mortality (6.3). The epidemiological significance of AII is determined by the rapid growth of this indicator: Mortality caused by AII grew by a factor of 32 since 1970.

Growth of mortality due to AII is not a consequence of an increase in registered morbidity, since the number of recorded AII cases in the country remained stable at 1.1 million per year throughout the period in question. At the same time, change in the etiological structure of AII was established: In addition to bacteria, viruses play a significant role (up to 70 percent) [3]. The course of AII of viral origin is relatively mild, in connection with which it may be supposed that registered cases of enteritis of viral origin are dominated by moderately severe and severe courses, often leading to death.

Septicemia is in next place (2.3 in 1987). The dynamics of mortality caused by it also exhibit an unfavorable trend. Considering the relatively high deadliness of infections in the purulent-septic group, it may be supposed that growth of mortality is a result of an increase in morbidity in this case. Factors responsible for the increase in frequency of purulent-septic infections include concentration of weakened individuals in large hospitals, the increasingly greater employment of parenteral procedures, growth of the role of conditionally pathogenic microorganisms, and development of hospital strains of disease agents distinguished by high virulence and stability in relation to environmental factors, and polyresistance to antibiotics [4].

A relatively high and continually increasing mortality is observed in relation to viral hepatitis (0.81 in 1970, and 2.2 in 1987), which is consistent with growth of viral hepatitis morbidity.

Meningococcal infection should also be named among the causes of death that are still highly significant. The intensive indicator for mortality caused by such infection is increasing, and it is approaching unity (0.8 in 1987). Replacement of disease agent groups plays a certain role in this situation besides the increase in morbidity. Thus according to Demina et al. [2], an increase in the etiological role of serological group B

meningococci has been noted in recent years, especially among young children, eliciting the most severe cases of illness.

As far as the rest of the nosological forms in the class of infectious and parasitic diseases are concerned, mortality due to them does not exceed 0.5. The highest mortality recorded in this disease group is associated with measles (0.34 in 1987) and salmonellosis (0.32).

Mortality due to infections which have been fought in the last decades by preventive immunization of the corresponding population groups is still lower (within 0.02-0.07). They include tetanus, diphtheria, whooping cough and poliomyelitis. The lowest mortality (0.01 and lower) is recorded in relation to rabies, brucellosis, scarlet fever and anthrax.

Thus unfavorable mortality trends in the period in question are associated with infectious diseases which are characterized by the most intensive indicators and which make up 57 percent of the structure of mortality caused by class I diseases.

In the rural population, mortality associated with the nosological forms of disease examined above exceeds the corresponding indicators for the urban population as a rule. This ratio is especially dramatic (a three- to sixfold difference) in relation to viral hepatitis and AII.

Mortality was examined from the aspect of age in order to reveal the groups with the highest risk.

Comparison of the age structure of persons dying of all causes and of class I diseases revealed and confirmed a known feature of infectious pathology—a shift in the proportion in the direction of younger age groups. Thus while persons dying of all causes in 1987 were 7.6 percent children (up to 14 years old) and 67 percent were elderly (60 years and over), among persons dying from infectious and parasitic diseases these indicators were 53.2 and 14 percent respectively. It should be emphasized that the proportion of children (up to 14 years old) in the structure of mortality due to class I diseases has been increasing continuously in recent decades—from 15 percent in 1970 to 53.2 percent in 1987.

An analysis of age-related mortality caused by infectious and parasitic diseases (Table 2) revealed abrupt growth of mortality among children up to 1 year old—from 124.2 in 1970 to 401.5 in 1987.

**Table 2. Age-Related Mortality Caused by Infectious and Parasitic Diseases in the USSR (per 100,000 Population of the Corresponding Age)**

Age, Years	Year								
	1970	1979	1981	1982	1983	1984	1985	1986-	1987
Up to 1	124.2	348.5	459.0	450.1	469.1	444.1	419.9	-393.4	401.5
Up to 4	-	92.6	128.2	127.2	143.2	128.6	123.0	117.4	116.7
3-14	4.3	3.4	2.7	3.9	3.6	3.5	3.2	3.1	3.2
15-19	3.6	2.8	2.7	2.9	2.8	2.7	2.6	2.5	2.4
20-29	19.6	12.3	12.5	12	6.2	6.4	6.4	5.9	6
30-39	23.3	14.2	13.1	12.4	12	12.7	12.3	10.4	10.2
40-49	35.5	24.3	22.2	21	20.6	22	20.9	17.8	17.2
50-59	49.7	40.6	27.2	27.8	27.4	26.6	26.2	21.7	-21.6
60-69	70	36.9	30	27.5	26.3	25.2	25.1	20.8	21.2
70 and older	71.6	38.1	31.5	29.3	26.1	25.9	25.2	22.4-	21.8
Total for class I	26.6	22.9	24.4	24.0	25.1	24.0	23.3	21.2	-21.2

Another typical feature is the stable and consistent decrease in mortality indicators for all age groups, with the exception of children up to two years old.

These changes are responsible for the transition of the U-shaped curve most frequently encountered in relation to infectious diseases to an L-shaped age-related curve. Such a transformation is interpreted by demographers as evidence of extremely unfavorable dynamics of age-related mortality [1].

The highest mortality in 1987 (per 100,000 population of the corresponding age) among children up to one year old is recorded in relation to AII (233.7), septicemia (78.4), viral hepatitis (30.5), meningococcal infection (22.4), salmonellosis (12), measles (8.9) and shigellosis (4.6).

With further increase in age, mortality associated with all nosological forms decreases, and beginning at age 15 it does not exceed 1.0 per 100,000 population of the corresponding age. Mortality caused by tuberculosis is an exception: It increases with age, attaining a maximum among persons over 50 years old (16.2-18.2).

Consequently just six infectious diseases (AII, viral hepatitis, septicemia, meningococcal infection, salmonellosis, measles) are responsible for more than 96 percent of the country's child mortality. In this situation, the country's epidemic control and therapeutic services must focus their efforts on developing effective measures to prevent and treat these diseases, and at implementing them as quickly as possible.

Analysis of mortality in relation to the different republics revealed a sharp contrast in levels. In 1987, given an average unionwide mortality of 21.2 (per 100,000 population) due to class I diseases, the highest mortality was recorded in the Central Asian republics (58.1-86.2),

while the lowest was in the Baltic republics and the Belorussian SSR (7.3-10.4).

Analysis of mortality dynamics in relation to the class of infectious and parasitic diseases in 1970-1987 permits the assertion that the decrease in the unionwide indicator noted above (from 26.6 to 21.2) occurred due to the RSFSR, the BSSR, the UkSSR, and the Baltic and Transcaucasian republics. At the same time, mortality dynamics were characterized by growth in the Uzbek, Kirghiz, Tajik and Turkmen SSR—that is, in the republics with the highest indicators today.

Special mention should be made of mortality among children up to one year old due to infectious and parasitic diseases, recorded in the Turkmen (1,453.1 per 100,000 children up to one year old), Tajik (1,169.2), Uzbek (1,042.1) and Kirghiz SSR (953.8). These figures exceed the corresponding indicator for the country by a factor of three or more.

As far as mortality caused by individual diseases among the most significant nosological forms is concerned, while it maintains the features noted above, it does vary within significant limits. Thus in 1987 the mortality indicators for the different republics varied by a factor of 1,284 in relation to AII's of indefinite etiology, by a factor of 724 in relation to measles, by a factor of 47 in relation to viral hepatitis, by a factor of 38 in relation to meningococcal infection, by a factor of 34 in relation to shigellosis, and by a factor of 22 in relation to septicemia. This is evidence both of pronounced differences in conditions promoting the onset of death as a result of these illnesses, and of the existence of significant reserves for reducing mortality.

This information on population groups, regions and nosological forms that are most unfavorable in relation to mortality caused by infectious diseases supplements the general description of the epidemic condition of the

population, and determines the high significance of infectious pathology. It can promote selection of better ways of preventing and fighting infectious diseases.

#### Conclusions

1. Infectious diseases are not a cause of the growth of overall mortality recorded in the country. Among the causes responsible for overall mortality in the country, infectious and parasitic diseases have occupied a stable 2-3 percent over the last several decades, while mortality due to class I diseases is on the order of 21 per 100,000 population.

2. The main causes of death due to infectious diseases are tuberculosis (37 percent), acute intestinal infections (30 percent), septicemia (11 percent), viral hepatitis (11 percent), meningococcal infection (4 percent), measles (2 percent) and salmonellosis (1.5 percent).

3. Almost half of all persons dying due to infectious and parasitic diseases (48 percent) are children up to two years old.

4. The highest levels are continually recorded in relation to all infections in the republics of Central Asia, while the lowest are in the Baltic republics and the BSSR.

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### Synthesis and Pharmacodynamics of 2- and 6-Substituted [Pyrid-3-yloxy]acetic Acid Derivatives

917C0245A Moscow

*KHIMIKO-FARMATSEVTICHESKIY ZHURNAL*  
in Russian Vol 24 No 9, Sep 90 (manuscript received  
25 Sep 89) pp 28-32

[Article by T. A. Voronina, O. M. Glozman, E. K. Orlova, V. I. Kuzmin, L. D. Smirnov, T. L. Garibova, I. V. Khromova, W. Sauer, A. Rostok, and H. Sigemund, Scientific Research Institute of Pharmacology, USSR Academy of Medical Sciences, Moscow; Hermed National Pharmaceutical Enterprise, Main Administration, Arzneimittelwerk, Dresden, GDR]

UDC 615.214.31.012.1

[Abstract] A series of 2- and 6-substituted pyrid-3-yloxyacetic acid congeners were synthesized and tested for nootropic properties in comparison with meclofenoxate. Trials on outbred albino 16-20 g male mice showed that certain of the compounds were superior to piracetam and equivalent to meclofenoxate as nootropics. For example, 2-dimethylaminoethyl ester (2-ethyl-6-methylpyrid-3-yloxy)acetic acid (I) and 2-dimethylaminoethylamide (2-ethyl-6-methylpyrid-3-yloxy)acetic acid dioxalate were shown to improve memory, (2-ethyl-6-methylpyrid-3-yloxy)acetic acid 3-dimethylaminopropyl ester displayed antihypoxic properties (II), and I and II also exerted tranquilizing action. Sedative or myorelaxant complications were not observed; however, these agents have been shown to be more toxic than piracetam and meclofenoxate. Tables 3; references 10: 6 Russian, 4 Western.

### Synthesis and Pharmacodynamics of Enamidine and 1,4-Dihydropyridine Derivatives

917C0245B Moscow

*KHIMIKO-FARMATSEVTICHESKIY ZHURNAL*  
in Russian Vol 24 No 9, Sep 90 (manuscript received  
3 Nov 89) pp 32-35

[Article by Ye. Yu. Murugova, O. B. Romanova, L. M. Alekseyeva, Ye. A. Rumyantsev, I. F. Fayermark, G. Ya. Shvarts, and V. G. Granik, Chemical Pharmaceutical Scientific Research Institute imeni S. Ordzhonikidze, Moscow]

UDC 615.277.3+615.281.8]:547.759.32]:012.1

[Abstract] A series of enamidine and 1,4-dihydropyridine derivatives were synthesized for multi-animal screening studies. The results showed that certain derivatives of 3-( $\alpha$ -N,N-dimethylaminoethylidene)amino-cyclohexen-2-one-1 (I) and 3,4-diethoxycarbonyl-1,4,5,6,7,8-hexahydroquinolone-5 were weakly hypotensive. In addition, several derivatives of compound I were moderately anti-inflammatory and analgesic. It appears that in some cases the effects may have

been due to calcium blocking mechanisms. None of the compounds exceeded nifedipine or amidopyrine in their pharmacodynamic spectrum. The LD<sub>50</sub> values for these agents ranged from 350 to > 1000 mg/kg. Tables 3; references 12: 4 Russian, 8 Western.

### Synthesis and Antiviral Activity of 6-Nitro-7-Oxo-4,7-Dihydroazolo[5,1-C][1,2,4]Triazines

917C0245C Moscow

*KHIMIKO-FARMATSEVTICHESKIY ZHURNAL*  
in Russian Vol 24 No 9, Sep 90 (manuscript received  
30 Jan 89) pp 41-44

[Article by V. L. Rusinov, Ye. N. Ulomskiy, O. N. Chupakhin, M. M. Zubairov, A. B. Kapustin, N. I. Mitin, M. I. Zhiravetskiy, and I. A. Vinograd, Ural Polytechnical Institute imeni S. M. Kirov, Sverdlovsk]

UDC 615.281.012.1.07

[Abstract] A series of 6-nitro-7-oxo-4,7-dihydroazolo[5,1-c][1,2,4] triazines (I) were synthesized for screening as an antiviral agent. Trials on chicken fibroblast cell cultures infected with Aujeszky virus led to the identification of the sodium salts of 2-methylpyrazolotriazine of I and the dimethylammonium salt of 6-nitro-7-oxo-4,7-dihydro-1,2,4-triazolo[5,1-c][1,2,4]triazine as the most potent antiviral agents. Tissue culture toxicity testing showed that addition of an alkyl fragment to the triazine nitrogen atom markedly increased toxicity. In general, toxicity was directly related to hydrophobicity of the compounds. Tables 2; references 14: 7 Russian, 7 Western.

### Synthesis and Antiviral Activity of Hydrophosphoryl Inosine Derivatives

917C0245D Moscow

*KHIMIKO-FARMATSEVTICHESKIY ZHURNAL*  
in Russian Vol 24 No 9, Sep 90 (manuscript received  
3 Oct 89) pp 44-48

[Article by V. V. Belakhov, A. A. Levina, Yu. D. Shenin, B. I. Ionin, Ye. B. Shtilbans, L. A. Rachkovskaya, E. V. Chekunova, S. S. Marennikova, and M. A. Shenyder (dec.), All-Union Scientific Research Technological Institute of Antibiotics and Enzymes with Medical Applications, Leningrad]

UDC 615.281:547.241].012.1.07

[Abstract] Data are presented on the synthesis and antiviral screening of a series of derivatives of 9-[3'-hydroxy-4'-alkyl (aryl)-5'-(P-hydroxy-P-hydrophosphoryl)-6'-hydroxymethyl-1,4'-mor-pholyl]hypoxanthine (I), an inosine derivative. Testing of the compounds on 10- to 11-day chick embryos showed that the compounds were active against vaccinia virus, Rous sarcoma virus, and A and B influenza viruses. In conjunction with low toxicity, the

activity of these compounds against RNA and DNA viruses suggests that modification of inosine dialdehyde with primary amines and phosphorylation is a promising avenue for synthesis of novel antiviral agents. Tables 4; references 14: 8 Russian, 6 Western.

#### Synthesis and Antiviral Activities of Phosphorylated Diaza-18-Crown-6

917C0245E Moscow

KHIMIKO-FARMATSEVTICHESKIY ZHURNAL  
in Russian Vol 24 No 9, Sep 90 (manuscript received  
5 Oct 89) pp 51-53

[Article by N. G. Zabiroy, O. K. Pozdeyev, V. A. Shcherbakova, T. N. Shumilova, G. Kh. Gilmanova, and R. A. Cherkasov, Kazan University imeni V. I. Ulyanov-Lenin]

UDC 615.281:547.898].012.1

[Abstract] Trials with phosphorylated derivatives of diaza-18-crown-6 (I) showed that while the starting compound lacked antiviral activity, certain of the derivatives were active. Highest activity against influenza H3N2 viruses was displayed by alkaline salt complexes of N-phosphorylated (thio)amides and bis(thio)ureas of compound I in *in vivo* studies on 16-18 g mice. The survival rate of the treated animals rose from 24.1 for compound I to 70.0-89.7 percent with the various derivatives, with the protection index attaining 53.7-79.4 percent. The results were less impressive in studies on infected chorioallantoic membranes, but indicate that phosphorylation of diaza-18-crown-9 ethers may yield valuable antiviral agents. Tables 2; references 5: 4 Russian, 1 Western.

#### Effect of Antioxidants on Hemoglobin Oxidation in Presence of Phospholipid Dispersions

917C0360A Moscow

KHIMIKO-FARMATSEVTICHESKIY ZHURNAL  
in Russian Vol 24 No 2, Dec 90 (manuscript received  
13 Dec 89) pp 7-9

[Article by M. A. Sablina, I. P. Ushakova, Ye. I. Zakharova, G. A. Serebrennikova, S. M. Alekseyev, K. A.-V. Shuaipov, and R. P. Yevstigneyeva, Moscow Fine Chemical Technology Institute imeni M. V. Lomonosov]

UDC 615.362.111.11.014.6:577.352.2].014.425

[Abstract] The effect of a number of antioxidants on hemoglobin oxidation in the presence of phospholipid dispersions was studied as part of an ongoing investigation of hemosomes (lipids with encapsulated hemoglobin) which are a convenient erythrocyte model and which may be used in researching lipid-protein interaction. The antioxidants involved were  $\alpha$ -tocopherol and its analogs which differ in the side isoprenoid chain at position 2 of the chroman nucleus. The results demonstrated that the antioxidants effectively lengthen the

period of hemoglobin functional activity in the presence of phospholipid dispersions using approximately 3 mass percent of antioxidant to the amount of lipid material. In addition, it was shown that the antioxidants  $\alpha$ -tocopherol and derivative V (side chain of  $(CH_2)_{18}CH_3$ ) increased the period of hemoglobin functional activity from one day (without the antioxidant, vesicular dispersion of egg phosphatidylcholine) to 28 and 30 days respectively, while the control solution, which was not exposed to lipids, was functional for 45 days. These findings suggest that lengthening the side chain at position 2 of the chroman nucleus evidently results in more effective inhibition of hemoglobin in the presence of phospholipid dispersions by means of lipid peroxidation products. Finally, the results demonstrate that phospholipids with unsaturated fatty acid radicals may be employed with the use of antioxidants in producing hemosomes. Figures 1; tables 1; references 9: 6 Russian, 3 Western.

#### Isoquinoline Derivative Synthesis and Antiaggregant and Hypotensive Activity

917C0360B Moscow

KHIMIKO-FARMATSEVTICHESKIY ZHURNAL  
in Russian Vol 24 No 12, Dec 90 (manuscript received  
5 Oct 89) pp 22-24

[Article by M. Yu. Dormidontov, B. Ya. Syropyatov, R. Z. Dautova, B. B. Aleksandrov, V. S. Shklyayev, M. I. Vakhryn, and A. G. Mikhaylovskiy, Organic Chemistry Institute, Ural Department, USSR Academy of Sciences; Perm Pharmaceutical Institute]

UDC 615.273.52.615.225.2]:547.833.1].07

[Abstract] Since the onset of vascular thrombosis in cases of hypertension frequently exacerbates cardiovascular disease, the development of a drug with both antiaggregant and hypotensive properties would be of great benefit. Accordingly, such a substance was sought for among isoquinoline derivatives. The results demonstrated that compounds with a methoxy group at positions 6 and 7 of the isoquinoline ring as well as an amino group and benzyl radical favor the expression of high antiaggregant activity. In addition, it was shown that compounds with an amino group and those similar to papaverine are hypotensive. The findings indicated that 3,3-dimethyl-6,7-dimethoxy-1-(2-dimethylaminoethyl)-3,4-dihydroisoquinoline, trimethyl[2-(3,3-dimethyl-6,7-dimethoxy-3,4-dihydroisoquinolinium-1)]-ethylammonium diiodide, and 1-(R<sup>3</sup>-methyl)-3,3-dimethyl-6,7-dimethoxy-3,4-dihydroisoquinolines have the best combination of antiaggregant and hypotensive activity. Tables 3; references 8: 7 Russian, 1 Western.

**Synthesis and Antiviral Activity of Substituted Piperidines and Perhydroquinolines**

917C0360C Moscow

*KHIMIKO-FARMATSEVTICHESKIY ZHURNAL*  
in Russian Vol 24 No 12, Dec 90 (manuscript received  
19 Jan 90) pp 27-29

[Article by P. V. Reshetov, A. P. Krivenko, Ye. I. Boreko, G. V. Vladyko, and L. V. Korobchenko, Saratov University imeni N. G. Chernyshevskiy; Belorussian Epidemiology and Microbiology Scientific Research Institute, Minsk]

UDC 615.281:578.8]:[547.822.3+547.831].012.1

[Abstract] As part of the continuing search for novel highly effective antiviral preparations, a number of phenyl- and alkyl-substituted piperidines and perhydroquinolines were synthesized and their antiviral activity was investigated. These compounds were produced by liquid phase catalytic hydromethylation of 1,5-diketones and pyrylium salts. The antiviral activity was tested on tissue cultures infected with smallpox vaccine, herpes simplex I, classical fowl plague, vesicular stomatitis, Venezuelan equine encephalitis, Echo, and respiratory syncytial viruses. As a result, two substances were found to be effective against smallpox vaccine viruses, one of which—2,6-diphenyl-N-methylpiperidine—was effective at a wide range of concentrations and non-toxic. These findings suggest that the search for potential antiviral agents among azaheterocycles with similar structures should be continued. Tables 3; references 6: 1 Russian, 5 Western.

**Synthesis of Novel Antiarrhythmia Agent: Bonnecor**

917C0360D Moscow

*KHIMIKO-FARMATSEVTICHESKIY ZHURNAL*  
in Russian Vol 24 No 12, Dec 90 (manuscript received  
27 Apr 90) pp 51-53

[Article by A. P. Skoldinov, A. N. Gritsenko, Kh. Vunderlikh, A. Shtark, Ye. Karstens, and D. Loman, Pharmacology Scientific Research Institute, USSR Academy of Medical Sciences, Moscow; NP [as published] Artsnaysmittelwerk, Dresden]

UDC 615.22:547.869.2].012.1

[Abstract] Bonnecor (3-carbethoxyamino-5-dimethylaminoacetyl-10,11-dihydro-5H-dibenz[b,f]azepine chlorhydrate) is synthesized from 3-amino-5-acetyldibenzazepine, which is heated with a propanol solution of potassium hydroxide. This product is then converted into 3-carbethoxyaminodibenzazepine by means of ethyl ether of chloracetic acid. Further reaction with chloracetyl chloride yields 3-carbethoxyamino-5-chloracetyldibenzazepine, which when reacted with dimethylamine and then purified produces bonnecor. Bonnecor has been shown to be

stable in light and is not oxidized in air. Results of tests in 10 clinics in the USSR and GDR have demonstrated that it is highly effective in treating various forms of arrhythmia. Bonnecor also has been shown to decrease fibrillation and has been approved for cardiological use in both countries. It is manufactured in tablet and ampule form. References 7: 3 Russian, 4 Western.

**Testing for Emoxypine With Membrane Electrode**

917C0360E Moscow

*KHIMIKO-FARMATSEVTICHESKIY ZHURNAL*  
in Russian Vol 24 No 12, Dec 90 (manuscript received  
14 Mar 89) pp 77-78

[Article by N. M. Kocherginskiy, N. V. Shvedene, and A. A. Shvedova, Chemical Physics Institute, USSR Academy of Sciences, Moscow]

UDC 615.272.014.425.017:615.273.52].07:543.257.1

[Abstract] A technique based on the use of a membrane electrode was proposed as a means of direct potentiometric determination of emoxypine concentration. The potentiometric measurements are performed with a digital ionometer. The membrane electrode is a cylindrical teflon shell filled with a standard emoxypine solution. Results of testing demonstrated that this membrane electrode is suitable for measuring the emoxypine concentration in aqueous solutions. Figures 2; references 5: Russian.

**Novel Polysynthetic Heart Glycosides and Their Biological Activity**

917C0409A Tashkent KHIMIYA PRIRODNYKH

*SOYEDINENIY* in Russian No 6, Nov-Dec 90  
(manuscript received 15 Jan 90) pp 776-779

[Article by I. F. Makarevich, I. S. Terno, T. V. Slyusarskaya, S. N. Petrova, L. Ya. Topchiy, and A. A. Shepel; All-Union Scientific Research Institute for the Chemistry and Technology of Medicines, Kharkov; Kharkov Pharmaceutical Institute]

UDC 615.224:547.918

[Abstract] With the aim of studying the relationship between chemical structure and biological activity, digitoxigenin-3B-O-B-D-xylopyranoside (I) and digitoxin-4''-O- $\alpha$ -L-rhamnopyranoside (II), novel heart glycosides, were synthesized on the basis of digitoxigenin and digitoxin. The glycosides were synthesized by the Kengigs-Knorr method with the V. T. Chernobaya modification. A mixture of mercuric oxide and silver carbonate was used as the HBr acceptor. It was established that the compounds exhibited relatively high biological activity where compound I was 1.7 times more active than digitoxin and compound II was 1.4 times more active than purpureaglycoside. Although located far from the aglycone, the L-rhamnose ring had a significantly greater effect on biological activity than the D-glycoside ring, which was located at the same distance. Figures 1; references: 3 Russian.

**Metabolism of Calcium, Vitamin D and Enzymes of Metabolism of Xenobiotics During Chronic Action of Mycotoxins**

917C0224 Moscow VOPROSY PITANIYA in Russian  
No 5, Sep-Oct 90 (manuscript received 21 Apr 89)  
pp 25-30

[Article by I. N. Sergeyev, N. M. Piliya, Ye. E. Kuzmina et al.; Institute of Nutrition; USSR Academy of Medical Sciences; Moscow]

616-008.924.1+616-008.931:577.161.2]-02:615.9]-07

[Abstract] A study of the effect of prolonged ingestion of low doses of toxin T-2, desoxynivalenol and aflatoxin B<sub>1</sub> on activity of enzymes of foreign compounds, including the toxins themselves, on calcium homeostasis and the metabolism and reception of vitamin D in rats involved experiments on 176 +/- 8 g male Wistar rats. The rats received a semisynthetic ration containing 0.6 percent of calcium and 0.6 percent phosphorus and the minimum recommended vitamins. There were four groups of rats: 1-control group, 2-rats receiving toxin T-2, 3-rats receiving desoxynivalenol, 4-rats receiving aflatoxin B<sub>1</sub>. Non-toxic doses of trichotetsen mycotoxins which matched the real level of contamination of food were injected into the rats in subacute and subchronic experiments. Rats received T-2 toxin (0.063 mg/kg), desoxynivalenol (1.6 mg/kg) and aflatoxin B<sub>1</sub> (0.008 mg/kg) for six months. Metabolic enzymatic activity of the foreign substance in the liver and small intestine mucosa changed moderately. All mycotoxins produced weak hypocalcemia. The ionized calcium concentration in blood serum decreased only after use of T-2 toxin and was accompanied by an increase of the PTH level. Alkaline phosphatase activity and calcium transport in the small intestine were practically unchanged. T-2 toxin decreased the concentration of 25-OH-D in the blood serum and 25-hydroxylase activity in the liver. Formation of 1.25(OH)<sub>2</sub>-D<sub>3</sub> and 24.25(OH)<sub>2</sub>D<sub>3</sub> did not change significantly. T-2 toxin inhibited regulatory changes in 1-hydroxylase 25-OHD<sub>3</sub> activity due to the activity of PTH and the cyclase activator forskolin. Prolonged action of low doses of toxin T-2, desoxynivalenol and aflatoxin B<sub>1</sub> caused moderately pronounced changes of activity of enzymes of metabolism of foreign compounds in the liver and small intestine mucosa. The changes are adaptive changes. The mycotoxins, especially trichotetsin, may impair calcium metabolism and its regulation. This must be considered when calculating the potential danger to man. References 17: 8 Russian, 9 Western.

**Concerning Disturbances of Contractile Capacity and Capillary Blood Supply of Heart Muscle Under Conditions of Effect of Toxic Doses of Catecholamines and Their Pharmacological Correction**

917C0225 Yerevan KROVOBRASHCHENIYE in Russian Vol 23 No 2 Nov-Dec 90 (manuscript received 15 Sep 88) pp 3-6

[N. F. Krasnikov, R. Sh. Matevosyan and S. A. Sisakyan; Yerevan Medical Institute]

UDC 616.127+616.16:615.35

[Abstract] The study involved experiments on 36 spontaneously contracting explants of embryonal myocardium of chicks of a 6 to 7 day incubation in a culture. Adrenalin, isadrin, verapamil and intal were dissolved in the nutrient medium in specific concentrations and then introduced into the culture medium of the contracting explants. A separate series of experiments measured the effects of adrenalin (2 mg/kg), isadrin (5 mg/kg) and intal (1 mg/kg) on the capillary blood supply in dynamics. Isadrin and adrenalin showed similar  $\beta$ -adrenomimetic activity but adrenalin produced lesser chronotropic effect. The chronotropic effect and arrhythmogenic effect of intal resembles the effect of isadrin and adrenalin, indicating a  $\beta$ -adrenomimetic component in its action. Injection of adrenalin in a 2 mg/kg dose caused abrupt worsening of the myocardium capillaries blood supply due to narrowing of the capillaries and decrease of their functioning number because of its  $\alpha$ -adrenomimetic effect. Injection of isadrin against a background of the action of adrenalin after one hour increased the overall length of capillaries by 20 percent, the metabolic surface by 43 percent and the capacity of the capillary channel by 69 percent due to an increase of functioning capillaries by 20 percent and their diameter by 18 percent. Rats receiving intal against a background of adrenalin showed the drug's effect only after three days. The vasodilative effect of intal seen after three days after adrenalin injection was not due to its  $\beta$ -adrenomimetic effect. It may be due to the ability to inhibit the lipids peroxide oxidation activity. Figures 1; references 4: Russian.

**Neurotrophic Factors, Isolated From Central Nervous System**

917C0347 Moscow USPEKHI FIZIOLOGICHESKIKH NAUK in Russian Vol 21 No 4, 1990 pp 138-142

[Article by G. N. Akoyev and N. I. Chalisova]

[Abstract] A survey of the literature was used to discuss the possibility of the presence of nerve growth factor in the central nervous system, the possible pathway by which neurotrophic factors found in the central nervous system affect peripheral neurons, the effect of cutting central processes of sensitive neurons of animals in the neonatal period on the state of neurons of spinal ganglia. Transport of neurotrophic substances may possibly play an important role in establishing connections between neurons in embryogenesis for formation of functional systems and for maintaining the structural integrity of the nervous system in adult animals. Possibly only those neurons of the central nervous system survive in the process of embryogenesis which establish adequate connections with peripheral tissues. It is possible that, in the limits of one functional system, neurons are associated transynaptically with the aid of a common chemical language, neurotrophic factors. References 28: 4 Russian, 24 Western.

**Synthesis of Endorphins in Rat Lungs, Heart, Kidneys and Testes May be Regulated by Glucocorticoids and Dopamine**

917C0392 Moscow DOKLADY AKADEMII NAUK SSSR in Russian Vol 314 No 6, Dec 90 (manuscript received 15 May 90) pp 1503-1506

[Article by A. D. Dmitriyev, Ye. A. Kizim and M. B. Smirnova; Institute of Higher Nervous Activity and Neurophysiology; USSR Academy of Sciences; Moscow]

UDC 612.17.46.616

[Abstract] A study of the possibility of regulating endorphins synthesis by glucocorticoids in the rat heart, lungs, kidneys and testes involved experiments on male Wistar rats weighing 270-300 g. Experimental group rats received dexamethasone (0.2 mg/rat) or haloperidol (0.6 mg/rat) injections daily for six days while control group rats received physiological solution. Adrenalectomy of the rats produced a reliable increase of concentration of immunoreactive (ir)  $\beta$ -endorphins in the heart, lungs, kidneys and testes. A reliable increase of concentration of ir- $\alpha$  and ir- $\gamma$  endorphins occurred in the lungs and kidneys. Dexamethasone injections reliably decreased the ir- $\beta$ -endorphins concentration only in the heart and lungs and the ir- $\alpha$ -endorphins concentration in the lungs. Dexamethasone injections did not decrease the concentration of any of the endorphins in the testes but increased the concentration of ir- $\alpha$ -endorphins and ir- $\gamma$  endorphins in the kidneys in one experiment. Injection of the glucocorticoids decreased the proopiomelanocortin-peptides concentration in the anterior lobe of the hypophysis and their concentration increased after adrenalectomy. The studies showed that there is, in rat heart, lungs, kidneys and testes, groups of cells in which proopiomelanocortin-peptides formation is regulated by glucocorticoids and other groups of cells in which such formation may be regulated by dopamine. Both known

types of negative regulation of expression of the proopiomelanocortin gene, typical of the anterior lobe and typical of the intermediate lobe of the hypophysis, may occur for proopiomelanocortin-producing cells of rats internal organs. Figures 4; references 11: 1 Russian, 10 Western.

**Sporadic Cases of Legionella-Induced Pneumonia**

917C0397 Moscow KLINICHESKAYA MEDITSINA in Russian Vol 68 No 12, Dec 90 (manuscript received 17 Apr 90) pp 35-36

[Article by E. L. Shchennikov, V. N. Moiseyev, V. Ya. Uriyevskiy and L. G. Gridneva; Department of Propeutics of Internal Diseases (head - professor I. Z. Batkin); Khabarovsk Medical Institute; Khabarovsk Regional Clinical Hospital (Chief Physician - A. V. Kravets)]

UDC 616.24-002-022.7

[Abstract] Five cases of Legionella-induced pneumonia were diagnosed in 1988-1989 in the pulmonological department of Khabarovsk Regional Clinical Hospital and were confirmed serologically. The clinical picture of the disease resembled that of croupous pneumonia. Use of penicillin, cephalosporine and aminoglycosides was ineffective. In one case, the pneumonia progressed and spread during this antibiotic therapy. Erythromycin produced a rapid therapeutic effect after intravenous use in the first days of treatment and then in a dose of 2.0 g/day. Temperature dropped and intoxication decreased on the 3rd to 4th day. The clinical x-ray picture showed massive changes in the lungs. The illness was characterized by an acute onset, resembling that of croupous pneumonia; a severe prolonged course, semisegmentary type of infiltration with pleural involvement, a slight cough, absence of the usual cocci microflora in the sputum and bronchial washings, a pronounced shift of the peripheral blood to the left and pronounced increase of the erythrocyte sedimentation rate. Two case histories were presented. References 4: Russian.

### Conceptual Factors in Health Care Reform in Ukraine

917C0418A Kiev VRACHEBNOYE DELO in Russian  
No 11, Nov 90 pp 3-6

[Article by Yu. P. Spizhenko, Minister of Health, Ukrainian SSR]

UDC 614.2(477)

[Text] Health care in Ukraine, as in the rest of the Soviet Union, is undergoing extensive reorganization due to the impact of perestroika with all of its socioeconomic and societal ramifications. Problems with the system simply reflect negative aspects of Soviet society engendered by decades of stagnation.

Analysis of health care in Ukraine is difficult because of its plethora of problems. As some problems are eliminated new ones arise which require immediate attention, the Chernobyl disaster being a case in point. Consequently, reorganization of Ukrainian health care has become an urgent matter that demands a systematic, comprehensive approach. Reforms in Ukraine will have to be based on well-founded scientific principles in order to have any chance of success.

Health care professionals in Ukraine have not been inactive or neglectful. Much has been accomplished, but the fundamental problems generated by decades of absolute state control are not easy to overcome. Medical care, public health, and health administration are at a scandalously primitive level in Ukraine, and any reforms will have to be assessed in terms of their impact on overall morbidity.

Ukrainian demographic indicators of mortality and birthrate (on the order of 14-15) are worrisome. The average lifespan is 71 years: 66 years for men and 75 for women. The average annual population increase is on the order of 180,000 to 190,000, which is quite poor for a country of some 50 million people. Finally, Ukraine ranks in the 30th percentile among the industrially developed countries in terms of longevity, with no prospects for any improvements in the near future.

Health status indicators for children, adolescents, and working-age people—particularly women in childbearing years—take on a special significance since one fifth of the population consists of retirees. On the plus side is the fact that childhood mortality has been declining recently. Last year newborn mortality stood at 12.9 per 1,000 newborn, with levels of 10.0-12.0 reported in ten oblasts. These rather limited improvements are attributable to enhancements in neonatal care, despite the fact that social conditions in Ukraine have not changed for the better.

Preschool morbidity continues to be high, with 80 percent of the children reported to have health problems on entering the first grade. Epidemiologic data indicate that only 27 percent can be regarded as healthy. Furthermore,

33 and 47 percent, respectively, of the students at academic and vocational schools have been reported to suffer from neuropsychiatric disorders.

In a recent move the Ukrainian Ministry of Health has transferred school physicians to pediatric polyclinics and arranged for specialized training in order to improve the quality of medical care that preschoolers and schoolchildren receive.

Nevertheless, improvements in pediatric health and reduction of childhood mortality will depend to a large degree on mass screening of women, particularly those exposed to adverse work environments. Declining birthrate remains a constant worry since Ukraine has the lowest birthrate in the USSR.

Almost two million Ukrainian women work under conditions that pose a health risk. The incidence of extra-genital pathology and complications during pregnancy is on the rise and there are 4,000 cases of spontaneous abortion every year. It will take years to correct adverse ecological conditions responsible for poor health among women and children and which are responsible for congenital birth defects. It is the responsibility of the state sanitary services to remove women from work environments where they face a health risk. However, to date, they have failed to take a firm stance against industrialists who show more concern for production than for the health of their workers.

Despite creation of modern maternal and child health care centers, maternal mortality remains high at 38.2 cases per 100,000 births. In the Zaporozhye, Odessa and Kharkov oblasts this indicator stands at 66.5 due largely to medical incompetence. This is a situation that must be corrected.

Cardiovascular and respiratory disorders, malignancies and trauma account for most of the chronic, noninfectious health problems and are responsible for 93 percent of overall mortality.

The incidence of hypertension in Ukraine is 7,300 cases per 100,000 persons, reaching 11,000 in Vinnitsa, Khmelnytskyi and Cherkassy oblasts, and 12,000 in Zhitomir Oblast. The health departments in these oblasts are conducting active screening campaigns to uncover such cases. An objective criterion of the effectiveness of early diagnosis and treatment would be reduction in mortality from complications of hypertension.

In Odessa, Kiev and Kharkov oblasts most patients with hypertension have not been diagnosed, are not being treated, and mortality remains high and is on the increase. Prevention and treatment depend on accurate statistics and timely intervention. In addition, the incidence of the four primary causes of death reported abroad exceeds 4- to 5-fold the reported incidence in Ukraine. This means that we diagnose only 20-25 percent of the actual cases, a situation that requires immediate remediation.

Command-type administration and centralization, factors which inhibit initiative and efficient on-site problem-solving, are the primary reasons for unsatisfactory health care in Ukraine.

Fundamental reforms need to involve transition to the concept of health insurance, since only this approach to medical economics can offer any hope of alleviating our major problems. This means a change from strictly predetermined budgets to more flexible financing. Financing is expected to come from industrial enterprises, organizations, cooperatives, state financial institutions, foundations and from private individuals. This approach should result in a more stable financial base than that provided by revenues from taxes and ensure greater flexibility.

Transition to insurance-supported health care is going to be complicated and difficult because of current economic instability. The Ukrainian government is in the process of promulgating legislation dealing with health care and medical insurance to make such a changeover possible.

In addition, the population at large and the medical community will have to be re-educated to understand medical insurance, otherwise the system cannot be expected to succeed. Studies at the Kiev Scientific Research Institute of Social Hygiene and the Public Health Administration have shown that the reforms will provide medical personnel with social security within the framework of a market economy.

The reforms will entail the creation of general and personal insurance funds, implementation of fee-for-service medical practice, updating of obsolete equipment, and formation of medical cooperatives and associations. Some of the income will be used to guarantee free health care for the needy.

However, medical insurance is not a panacea. There will still be a shortage of equipment, drugs, supplies, and so forth. In addition, foreign experience has shown that medical insurance may have little impact on preventive health care, and additional problems may arise as a result of regional differences, particularly in rural areas.

Physician competence is another area of concern. Medical education will have to be improved, which will require a highly qualified faculty to produce competent graduates with a firm grasp of theoretical concepts. The present system is beset with poor teachers and an inadequate research record, especially in the basic sciences. Low admission standards, an uninterested student body,

unqualified teachers, and low academic demands serve to discourage the motivated students. Obviously, faculty quality must be raised to conform to international standards, and this will mean granting medical institutes a free hand in implementing the necessary reforms. Class time for social sciences will be reduced and time for clinical training will be increased. A standardized, computerized testing system will have to be established to monitor student progress. Finally, medical graduates will be required to pass a final qualifying examination in their specialty after graduation and every five years thereafter or lose their license to practice. New internship standards will have to be set. In the majority of foreign countries internships last from two to five years, depending on the specialty. The feeling at the Ukrainian Ministry of Health is that internships for surgeons, obstetricians and gynecologists, and emergency physicians should be two to three years in duration.

Medical reforms in Ukraine will involve transition to health insurance because of failure of the budgetary system to ensure adequate health care, alleviation of the most acute socioeconomic problems, systematic reforms and democratization of health delivery systems, use of economic incentives, changes in investing policies, and educational reforms at the medical school and postgraduate levels.

Success will depend on involvement and cooperation of specialists at faculties of social hygiene and public health, and health administration departments at scientific research institutes. In general, the entire spectrum of academic collectives will be involved in analysis and reforms of health delivery systems within the framework of new economic realities.

Production of medical equipment, supplies, instruments and drugs remains a problem in Ukraine and will not be solved by simply increasing funding. The ministry is developing a program that will encourage increased production of diagnostic equipment and expand the scope of available drugs. This includes joint ventures with foreign firms. Furthermore, special emphasis is placed on computerization of medical services, education, and administration.

Implementation of the Declaration of Ukrainian Sovereignty, Legislation on Economic Independence of Ukrainian SSR and the resolution shown by the Ukrainian Supreme Soviet and Ukrainian government on protecting the health of our nation will accelerate the pace and ensure success of these reforms. ©COPYRIGHT; Izdatelstvo "Zdorovya" "Vrachebnoye delo" 1991

**Plant Growth and Radiation Levels: Cytologic Assessment**

917C0246D Moscow TSITOLOGIYA I GENETIKA  
in Russian Vol 24 No 5, Sep-Oct 90 pp 33-37

[Article by P. K. Shkvarnikov]

UDC 575.24

[Abstract] Cytologic examinations were conducted on rootlet meristem cells of Kiyanka and Mironovskaya 808 wheat and rye plants collected within 30 km of the Chernobyl Nuclear Power Plant to assess the genetic sequelae of background radiation levels ranging from 180 to 300 mR/h. The results revealed that in the

experimental plants the percentage of cells with chromosomal abnormalities in rye were on the order of 2.666 percent, while the corresponding figures for the two varieties of wheat ranged from 1.158 to 2.561 percent. Furthermore, in the case of the wheat varieties the percentage of affected cells (1.075-3.187 percent) also was related to the biotype (awned, awnless, semiawned). These figures were 5.34-, 2.42- to 5.78-, and 4.07- to 6.71-fold greater than the corresponding statistical data for rye and wheat plants collected in areas unaffected by the nuclear fallout from the Chernobyl plant. The results obviously underestimate the actual rate of chromosomal aberrations since the study was limited to visually apparent changes. In addition, these observations underscore the potential human risk and the urgent need for expert teams to assess that neglected issue. Figures 2; tables 2.

**Diagnostic Antiserum Against Virulent *Yersinia***

917C0234A Moscow LABORATORNOYE DELO  
in Russian No 10, Oct 90 (manuscript received  
19 Dec 89) pp 66-68

[Article by I. V. Smirnov and V. I. Gorokhov, Chair of  
Microbiology, Ryazan Medical Institute imeni I. P.  
Pavlov]

UDC 616.98:579.842.23]-078.337

[Abstract] Antisera specific for *Yersinia* bearing 45-47 MD virulence plasmids were raised in chinchilla rabbits immunized with plasmid-bearing *Y. enterocolitica* 955P<sup>+</sup>. Following adsorption with a 955P<sup>+</sup> biomass the antisera were positive in slide agglutination tests only with *Y. enterocolitica* and *Y. pseudotuberculosis* strains bearing the virulence plasmids. Agglutination was evident within 15-30 sec using antisera diluted 1:20. The antisera did not agglutinate *Enterobacteriaceae* strains, retained their activity in liquid state at 4-8°C for at least one year, and have obvious utility as a diagnosticum specific for *Yersinia* sp. bearing virulence plasmids. References 5: 2 Russian, 3 Western.

**Capsule Formation by Melioidosis Pathogen in Organism**

917C0239A Moscow ZHURNAL MIKROBIOLOGII,  
EPIDEMIOLOGII I IMMUNOBIOLOGII in Russian  
No 9, Sep 90 (manuscript received 30 Oct 89) pp 6-10

[Article by B. I. Melnikov, S. F. Popov, A. I. Yakovlev et al.; Volgograd Scientific Research Anti-Plague Institute]

UDC 579.841.11:579.252/.233].083.1

[Abstract] A study of the possibility of formation of extracellular structure by *Pseudomonas pseudomallei* in the organism involved experiments on guinea pigs infected intraperitoneally by a daily agar culture of a virulent strain of *P. pseudomallei* C-41 in a dose of 2-3 billion microbial cells. Collection of peritoneal exudate by washing from the abdominal cavity by a buffered 0.15 M sodium chloride solution two and four hours after infection included material from five to six animals. Electron microscopic, immunocytochemical and electron-cytochemical examination revealed signs which characterize the observed, in vivo, extracellular formation of the melioidosis pathogen: clear-cut morphological outline, multistratified structure, impenetrability of it for specific immunoglobulins, ensurance of resistance to phagocytosis by preventing opsonization and sizes comparable to the diameter of a bacterial cell and showed that the pathogen can form a true capsule in the first hours of stay in the experimental animal's body. Figures 1; references 12: 7 Russian, 5 Western.

**Results of Clinical Tests and Prospects of Use in Medical Practice of Soviet Preparation of Human Recombinant Alpha-2 Interferon (Reaferon)**

917C0239B Moscow ZHURNAL MIKROBIOLOGII,  
EPIDEMIOLOGII I IMMUNOBIOLOGII in Russian  
No 9, Sep 90 (manuscript received 12 Aug 89) pp 61-67

[Article by Yu. T. Kalinin, A. A. Vorobyev, V. V. Bumyalis et al.; Main Administration "Biopreparat"; Ministry of Medical and Microbiological Industry; USSR; Moscow; Institute of Immunology; Ministry of Medical and Biological Industry; USSR; Moscow Oblast; 1st Moscow Medical Institute imeni I. M. Sechenov; Scientific Research Institute of Epidemiology and Microbiology imeni N. F. Gamaleya; Academy of Medical Sciences; USSR]

UDC 615.339:578.245].038

[Abstract] Results of clinical testing of reaferon and possible areas of its use were presented in two phases. The first phase of clinical testing determined the tolerance, reactogenicity and toxicity of reaferon and the second phase studied the safety, reactogenicity and therapeutic effectiveness of the drug when used in treatment of different oncological, infectious and other diseases. Reaferon proved to be safe and adequately tolerated. Side effects included body temperature increase for several hours after use, chills, sense of malaise, headache and dyspeptic disturbances. The moderate side effects depended directly upon the dosage used, disappeared after the 2nd-4th dose and did not require stoppage of use of the drug. Reaferon was quite effective in treatment of acute and chronic hepatitis B, different herpetic lesions of the skin and mucosa, viral meningoencephalitis, multiple sclerosis, smallpox, influenza, hairy cell leukemia, acute leukemias in children, breast cancer, Kaposi's sarcoma, reticulosarcomatosis and fungal mycosis. Results of clinical tests of the drug were presented in a lengthy table. References 7: 4 Russian, 3 Western.

**Diagnostic Effectiveness of Immunoenzymic Test-System for Determining Diphtheria Toxin Antibodies in Blood Serum During Clinical-Laboratory Test**

917C0239C Moscow ZHURNAL MIKROBIOLOGII,  
EPIDEMIOLOGII I IMMUNOBIOLOGII in Russian  
No 9, Sep 90 (manuscript received 29 Sep 89) pp 67-70

[Article by N. M. Nikityuk, L. Yu. Smurova, T. S. Shobukhova et al.; Scientific Research Institute of Standardization and Control of Medicinal Biological Preparations imeni L. A. Trasevich; USSR Ministry of Health; Moscow]

UDC 616.931-07:616.157-078.33

[Abstract] A commission study of the diagnostic values of an immunoenzymic test-system for detecting

diphtheria toxin in blood serum of diphtheria patients and persons suspected of having this infection involved examination of 240 blood serum samples from 209 persons making up an experimental and a control group. The experimental group included persons ranging in age from 18 to 60 years with different forms of diphtheria or tonsillitis and clinically healthy persons, suspected of having the disease. The test-system consisted of polystyrene plotting boards, a set of immunological reagents: F(ab)<sub>2</sub>-fragments of rabbit anti-diphtheria antibodies, a specific conjugate, healthy donor serum (negative control) and salts of buffer solutions. The test-system proved to be highly specific and extremely highly sensitive when blood was taken in the first days of the disease and when it was taken after use of therapeutic serum. Reproducibility of results was good. The high diagnostic effectiveness of the system justified recommendation of use of it in medical practice. Figures 5: Russian.

**Study of Effect of Different Yersinia Pestis Antigens on Cellular Link of Immunity**

917C0239D Moscow ZHURNAL MIKROBIOLOGII, EPIDEMIOLOGII I IMMUNOBIOLOGII in Russian No 9, Sep 90 (manuscript received 12 Aug 89) pp 85-89

[Article by I. V. Isupov, L. A. Nazarova, L. P. Pavlova et al.; All-Union Scientific Research Anti-plague Institute "Mikrob", Saratov]

UDC 615.371:579.843.95.015.4:612.438.017.1.076.9

[Abstract] A study of morphological and functional shifts occurring in thymus T-lymphocytes and cells of the mononuclear phagocytes systems in different lines and mongrel white mice after their immunization with live plague vaccine and antigens of the plague pathogen used microbiological, immunological and morphological methods. The live vaccine did not protect thymectomized mice from subcutaneous infection by the virulent strain. Live vaccine or individual Y. pestis antigens (fraction I) produced more pronounced functional and morphological activation of thymocytes and macrophages in C57BL/6 mice than in CBA mice. Y. pestis antigenic preparation fraction 1, fraction 2 and pesticin acted as T-cell mitogens and could cause in vitro proliferation of thymocytes. The thymocyte functional activity in mice increased significantly in the initial periods of immunogenesis under the effect of live vaccine or fraction 1. The number of T-suppressors in the thymus and spleen increased, especially after use of small doses of vaccine. The sensitivity of mice of different haplotypes to the injurious effect of the live vaccine was genetically determined. Fraction II decreased the lymphocytes level in the peripheral blood of the mice and destroyed the lymphocytes in their thymus and spleen. References 17: 13 Russian, 4 Western.

**A Highly Sensitive Method for Detection of Hepatitis A Virus Structural Proteins**

917C0329A Moscow LABORATORNOYE DELO in Russian No 12, Dec 90 (manuscript received 27 Dec 89) pp 77-78

[Article by G.K. Dzagurov, Yu. A. Kazachkov, Yu. Yu. Kusov, and F.S. Dzugkoyeva, Severo-Osetinsk Medical Institute, Ordzhonikidze; Institute of Poliomyelitis and Viral Encephalitis, USSR Academy of Medical Sciences, Moscow: "A Highly Sensitive Method for Detection of Hepatitis A Virus Structural Proteins"]

616.36-002-022:578.891]-078.337

[Text] Hepatitis A virus (HAV) belongs to the family of Picornaviridae and causes wide-spread intestinal infection. The protein composition of the virus has been studied insufficiently up to the present time. Previously, structural proteins of the virion VP1 (32-33 kD), VP2 (26-29 kD), and VP3 (22-27 kD) were observed in purified HAV preparations.<sup>1</sup> The protein VP4 (2.3 kD), which was predicted on the basis of the analysis of the primary structure of the HAV genome,<sup>3,4,8</sup> has not been detected up to this time.<sup>2,5,10</sup> The study of viral proteins is hampered because of the low production of HAV in cell cultures,<sup>1</sup> and, therefore, an increase in the sensitivity of protein analytical methods has become urgent. This communication reports the results of the development of a simple and highly sensitive method for detecting HAV structural proteins after electrophoresis in a polyacrylamide gel. For this, a number of modifications have been introduced into the well-known method for staining proteins with silver nitrate.<sup>7</sup>

The electrophoresis of proteins was conducted in gradient (12-17 percent) polyacrylamide gels with dimensions of 150 x 150 x 0.75 mm in a discontinuous buffer system in the presence of 0.1 percent sodium dodecyl sulfate.<sup>6</sup> All solutions for conducting electrophoresis and staining of the gel were prepared with deionized water.

After electrophoresis was completed, the gel was treated according to the following modus operandi.

1. The gel was fixed for 20 min in a solution containing 50 percent of methanol and 12 percent of acetic acid and then three times for 10 minutes in a mixture of 10 percent ethanol and 5 percent acetic acid.
2. The gel was held in 200 ml of an oxidizing solution: 0.0034 M K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>; 0.0032 N HNO<sub>3</sub>.
3. The gel was washed in several portions of deionized water until the gel was completely transparent.
4. The gel was placed in 200 ml of 0.012 M AgNO<sub>3</sub> containing 1 ml of 37 percent formaldehyde and 0.12 mg of benzotriazole and illuminated for 20 minutes by an incandescent lamp with a power of 0.5 kw.
5. Illumination of the gel being continued, the silver nitrate solution was removed, and a reducing solution

was added: 0.28 M  $\text{Na}_2\text{CO}_3$  containing 0.75 ml of 37 percent formaldehyde and 0.6 mg of benzotriazole per liter of solution. The first portion of reducing solution (100 ml) was removed after 10-20 seconds, and a second portion (300 ml) was added; colored bands of proteins appeared at this stage. The gel was placed in a 1 percent acetic acid solution in order to achieve the necessary degree of staining.

The *modus operandi* for staining presented above differs from the original method<sup>7</sup> in the following ways. The gel is washed more thoroughly from potassium bichromate than four times every 30 seconds (stage 2). A 1.5-fold greater amount of formalin was added to the reducing solution (stage 5). The gel was illuminated during stages 4 and 5, and not only during the first five minutes of stage 4, as recommended in the original method.

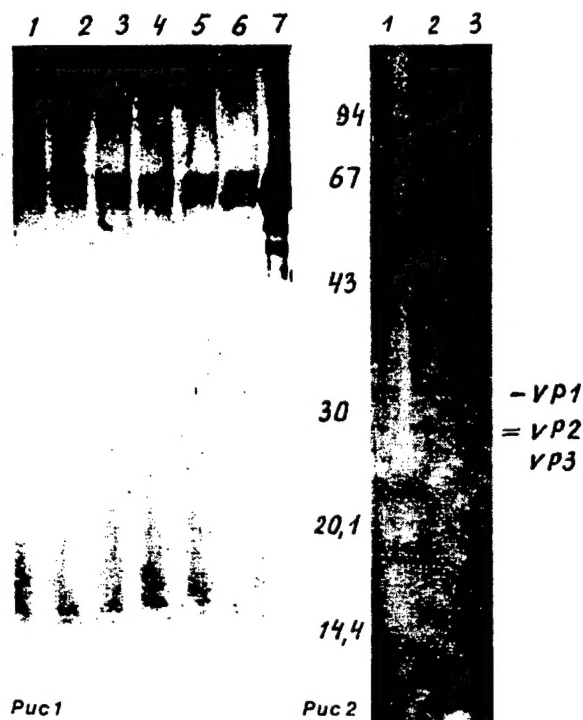
The sensitivity of the original method comprised only about 1 ng of protein in the band.<sup>7</sup> In Fig. 1 (See plate) the result of the staining of protein by the modified method is presented. Different amounts of ox serum albumin were subjected to electrophoresis and staining in accordance with the *modus operandi* presented above. The minimal detectable amount of protein comprised 0.1 ng in this case (track 3). Track 1 contained only the buffer for samples and served as a control of the staining.

The modified method was used to detect HAV structural proteins. The virus (strain HAS-15) was reproduced in an intertwined culture of kidney cells of a macaque rhesus monkey embryo (FRhK-4) and purified by means of centrifugation in a cesium chloride gradient.<sup>2</sup> The virus concentration of the preparation obtained was calculated starting from the titer of HAV particles determined by means of electron microscopy<sup>2</sup> and the known mass of Picornaviridae virus particles.<sup>9</sup> The result of electrophoretic analysis of the virus preparation is given in Fig. 2 (See plate). It can be seen that 1 ng of virus was completely sufficient to detect HAV structural proteins VP1, VP2, and VP3. The molecular masses of the proteins were determined by means of comparing their electrophoretic mobility with the mobility of a bank of marker proteins ("Pharmacia," Sweden) and comprised 33, 29, and 27 kD, respectively. That these proteins belonged to HAV structural proteins was confirmed by means of immunoblotting with a specific anti-HAV serum reconvalescent.

Thus, the modification of the procedure for staining proteins with silver nitrate proposed in this article increases the sensitivity of the method in comparison with the original<sup>7</sup> by at least one order of magnitude. This has made it possible to detect HAV structural proteins in a low concentration virus preparation. Due to the high sensitivity, the method can be used to detect HAV proteins and other viruses in clinical samples.

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**Figure 1.** Staining of ox serum albumin by silver nitrate after electrophoresis in polyacrylamide gel. 1—buffer for samples without protein; 2-7—0.01, 0.1, 1, 10, 100 and 1000 ng of albumin

**Figure 2.** Electrophoretic analysis of HAV proteins. 1-3—0.01, 0.1, and 1 ng of virus. On the left—position of marker proteins with an indication of molecular mass (in kD)

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